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EXPERIMENTAL NEPHROPATHIES

II. RENAL PHOSPHATASE AFTER POISONING WITH MERCURY BICHLORIDE, URANYL NITRATE AND POTASSIUM DICHROMATE

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CHICAGO

Phosphatase is an enzyme, first discovered in the mucosa of the duodenum by Grosser and Husler [1] in 1912, which splits the PO_4 ion from the esters of phosphoric acid. Two types have been differentiated, "alkaline," which is most active in a medium with p_H about 9.0, and "acid," which acts best in a medium with p_H between 5.0 and 6.0. Both types can be readily demonstrated in tissues by microtechnical (Gomori [2]) and chemical (Bodansky [3]) methods. This enzyme is present in blood serum (Bodansky [3]; Thannhauser and co-workers [4]), in bile (Freeman, Chen and Ivy [5]), in urine (Kutscher and Wolbergs [6]) and in human semen (Huggins and Johnson [7]).

The pattern of the distribution of phosphatase in the different organs of animals belonging to the same species is relatively constant (Gomori [2a]). Even in animals of different species the location of the enzyme in corresponding organs is much more constant than is its amount. Thus, MacFarlane, Patterson and Robinson,[8] using chemical methods, found increasing quantities of alkaline renal phosphatase in the following order of the species examined: rabbit, rat, guinea pig, dog, cat and mouse. Not only did the kidneys of the dog, the cat and the mouse contain more of the enzyme than did those of the rabbit, the rat and the guinea pig, but the renal phosphatase of the former group was activated to a much greater degree by magnesium. This observation is important in the selection of animals in which to study the phosphatase of the kidneys. Wherever found, alkaline phosphatase, stained by Gomori's method, is granular in appearance. In the kidneys it is limited to the brush border of the epithelium of the proximal convoluted tubules (Gomori [2a]; Hepler, Gurley and Simonds [9]). Elsewhere it is distributed through the cytoplasm of the cells. In the small intestine it is limited to the superficial epithelial cells of the mucosa.

Our experiments were undertaken with the hope of learning something concerning either the function or the mechanism of action of alkaline renal phosphatase. In another paper two of us [10] have presented evidence, confirming earlier work by Suzuki,[11] that in dogs potassium dichromate injures chiefly the first part, and mercury bichloride and uranyl nitrate chiefly the terminal portion, of the proximal convoluted tubule. In previous experiments we had determined the smallest dose of each of these poisons that would produce detectable injury of the renal tubules without visibly affecting the glomeruli. It appeared possible, therefore, that by injecting optimal doses directly into the blood stream varying degrees of damage up to complete necrosis might be inflicted on different parts of the proximal convoluted tubule and thus permit a study of these effects on the phosphatase activity of the kidneys.

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1. Grosser, P., and Husler, J.: Biochem. Ztschr. **39**:1, 1912.

2. Gomori, G.: (*a*) Proc. Soc. Exper. Biol. & Med. **42**:23, 1939; (*b*) J. Cell. & Comp. Physiol. **17**:71, 1941; Arch. Path. **32**:189, 1941.

3. Bodansky, A.: J. Biol. Chem. **101**:93, 1933.

4. Thannhauser, S. J.; Reichel, M., and Frattan, J. F.: J. Biol. Chem. **121**:697, 1937.

5. Freeman, S.; Chen, Y. P., and Ivy, A. C.: J. Biol. Chem. **124**:79, 1938.

6. Kutscher, W., and Wolbergs, H.: Naturwissenschaften **23**:559, 1935.

7. Huggins, C., and Johnson, A. A.: Am. J. Physiol. **103**:574, 1933.

8. MacFarlane, M. G.; Patterson, L. M. B., and Robison, R.: Biochem. J. **281**:720, 1934.

9. Hepler, O. E.; Gurley, H., and Simonds, J. P.: Proc. Soc. Exper. Biol. & Med. **44**:221, 1940.

10. Simonds, J. P., and Hepler, O. E.: Arch. Path. **39**:103, 1945.

11. Suzuki, T.: Morphologie der Nierensekretion unter physiologischen und pathologischen Bedingungen, Jena, G. Fischer, 1912.

METHODS

All our experiments were made on dogs, and the methods used are described in a previous paper.[10] One group of dogs was poisoned with mercury bichloride, a second group with potassium dichromate and a third with uranyl nitrate. The total volume of circulating blood was estimated in each animal by the formula: Weight in grams × 0.0925 = volume of blood in cubic centimeters. The poison was dissolved in isotonic solution of sodium chloride and injected slowly into a vein in such amount that the dog received one or more doses of 0.1 to 3 mg. of the poison for each 100 cc. of circulating blood. These dogs either died or were killed from twenty-eight hours to eight days after the first injection of the poison. The phosphatase of the serum and that of the renal cortex were determined by Bodansky's [9] method. Sections of the kidneys stained by Gomori's [2a] technic were employed in order to demonstrate any deviation from the normal in the amount, the form or the distribution of phosphatase in the tubular epithelium.

PHOSPHATASE IN NORMAL CONTROLS

Chemical determinations were made of the phosphatase in the blood serum of 13, and in the renal cortex of 37, unselected normal dogs, some of which had been subjected to acute experiments of various kinds in the department of physiology of Northwestern University Medical School. The results of these determinations are shown in table 1.

TABLE 1.—*Determinations of Renal and Serum Phosphatase in Normal Dogs*

	Dogs	Mean in Bodansky Units	Standard Deviation
Serum phosphatase.....	13	3.69 ± 0.32*	0.55 ± 0.23
Renal phosphatase.....	37	10.72 ± 0.33†	2.99 ± 0.23

* Per hundred cubic centimeters.
† Per gram.

The range of serum phosphatase was relatively narrow, as is shown by the comparatively small standard deviation; that of renal phosphatase was much wider, its standard deviation being about one fourth of the mean value. In some tissues, notably in bone, the amount of phosphatase is known to vary inversely with the age of the animal. No results appear to be available that would indicate whether this is true of the kidneys. Furthermore, prolonged ether anesthesia probably reduces the phosphatase activity of the kidneys. Since some of these animals had been subjected to such anesthesia for varying periods, this may have been a factor in the wide range of the number of phosphatase units for the different dogs in this series.

In sections of normal kidneys stained by Gomori's method the phosphatase, being sharply limited to the brush border of the epithelium, forms a narrow ring about the lumen of the proximal convoluted tubule. This characteristic distribution may be inherently related to the fact that it is the function of the epithelium of the proximal convoluted tubules to absorb the contents of the lumens and not to produce some substance and excrete it into the lumens.

PHOSPHATASE IN DOGS POISONED WITH MERCURY BICHLORIDE

Dogs were given mercury bichloride in the manner described under "Methods." The number and the size of the doses were varied for different animals so that they received one or more injections of 1, 2 or 3 mg., respectively, of mercury bichloride for each 100 cc. of blood. These animals died or were killed from twenty-eight hours to eight days after the first injection of the poison. The phosphatase of the serum was determined before, and from one to four times after, the injection, and the results in dogs with at least three determinations are shown in table 2.

TABLE 2.—*Determinations of Serum Phosphatase in Twelve Dogs Poisoned with Mercury Bichloride*

	Bodansky Units
Average of values before injection.....	3.00
Average of minimum values after injection....	2.75
Average of maximum values after injection....	4.75
Average of final values after injection.....	3.98

Since the averages in table 2 were obtained from a group of dogs not identical with that used in compiling table 1, there is a difference in the mean values of the controls in the two tables. The averages of the minimum and maximum values were obtained from the lowest and highest results, respectively, in the multiple determinations after administration of the poison. The average of the final values was calculated from the last determinations immediately before the animals were put to death. The average of the maximum values was about 58 per cent higher, that of the final values about 33 per cent higher and that of the minimum values less than 10 per cent lower than the average of the control values of the same animals before the injection of mercury bichloride. Poisoning of these dogs with this substance therefore produced definite alterations in the serum phosphatase during a period of at least eight days after the administration of the poison.

Changes in the phosphatase activity of the renal cortex are influenced by the amount of poison administered, as shown in table 3.

Of the 17 dogs used for the determinations in table 3, 6 died and 11 were killed five to eight days after the first injection of the poison. Of 6 dogs that were given in a single dose 3 mg. of mercury bichloride for each 100 cc. of blood, 5

died, and the remaining animal was killed five days after the administration of the bichloride. The kidneys of the 6 dogs in this group (table 4) contained an average of 7.62 Bodansky units of phosphatase per gram, which is 29 per cent below the normal mean. The dog that survived for five days had the highest renal phosphatase (11.31 units) in this group. The average of the values for renal phosphatase in 4 dogs that were given three injections of 1 mg. each of mercury bichloride for each 100 cc. of blood on successive days was practically normal (10.78 units, compared with the normal mean of 10.72 units). The average of the values for renal phosphatase activity in 6 dogs that received 2

TABLE 3.—*Determinations of Renal Phosphatase in Dogs Poisoned with Mercury Bichloride Showing Influence of Amount of Dose*

Dosage of $HgCl_2$	Dogs	Renal Phosphatase: Mean in Bodansky Units	Renal Phosphatase: Standard Deviation
3 mg. to 100 cc. of blood.....	10	9.10 ± 0.75	3.50 ± 0.53
2 mg. or less to 100 cc. of blood	7	16.57 ± 1.39	5.44 ± 0.97

TABLE 4.—*Determinations of Renal Phosphatase Showing Influence of Number of Injections of the Poison*

Dosage of $HgCl_2$ Mg.	Injections	Dogs	Average of Values in Bodansky Units	Range of Values in Bodansky Units
3	1	6	7.62	4.28 to 11.31
1	3	4	10.78	7.66 to 16.90
2	1	2	16.05	6.30 to 12.90 to 22.95
1	2	4		
1	1	1	19.05	

mg. of mercury bichloride for each 100 cc. of blood, either in single or broken doses, was approximately 50 per cent above the normal mean. One dog in this group died on the seventh day after injection of the poison, and its kidney had the lowest content of phosphatase (6.3 units) in the group. The kidneys of a dog that was given 1 mg. of mercury bichloride for each 100 cc. of blood yielded 19.05 units of phosphatase (77 per cent above the normal mean). The kidneys of another dog that received a similar dose gave such extremely high phosphatase values (27.12 units, more than two and a half times the normal mean) that it was not included in any of the calculations for this whole group of dogs.

The results of the determination of the phosphatase activity of the kidneys of this series of dogs by chemical means may be briefly summarized as follows: 1. A single dose of 3 mg. of mercury bichloride for each 100 cc. of blood was fatal to approximately 90 per cent of the animals within three days. 2. Such fatal doses produced extensive necrosis of the epithelium of the proximal convoluted tubules and materially reduced the phosphatase of the kidneys but did not completely destroy or inactivate the enzyme. 3. Smaller nonfatal doses of 2 mg. or less of mercury bichloride for each 100 cc. of blood seemed to stimulate the activity of the phosphatase of the kidneys, particularly when the dose was so small that microscopic examination revealed no actual necrosis of the tubular epithelium.

The results of the quantitative determinations of phosphatase in the kidneys of dogs poisoned with mercury bichloride were compared with sections of the same kidneys stained by Gomori's method. In normal canine kidneys the phosphatase is distributed in the brush border throughout the entire length of the proximal convoluted tubules, while the remainder of the cytoplasm of these cells is free. In the kidneys of dogs that received the minimum necrotizing dose of mercury bichloride the necrosis was limited to the distal half of the proximal convoluted tubules, particularly in their straight terminal portions, which lie in the outer part of the labyrinth along the medullary rays. The same kidneys showed no necrosis and little or no alteration in the intracellular distribution of phosphatase in the narrow aglomerular zone immediately beneath the renal capsule.[10] The earliest visible effect of necrotizing doses of mercury bichloride appears to be diffusion of the phosphatase throughout the cytoplasm. As long as the necrotic cell retains its form, its cytoplasm is diffusely stained by Gomori's method, but more palely than is the brush border of normal cells. The results of chemical analysis indicate that the pale staining is due to actual diminution in phosphatase activity and not merely to dilution by diffusion throughout the cell. In normal canine kidneys the lumens of the tubules do not contain detectable phosphatase. When cells of the proximal convoluted tubules disintegrate after damage by mercury bichloride, the detritus in the tubular lumens gives a positive phosphatase reaction by Gomori's method.

Figures 1*a* and 1*b* show serial sections from the kidney of a dog (K Hg 6) that received a single injection of 3 mg. of mercury bichloride for each 100 cc. of blood and died on the third day thereafter. In section *a*, stained with hematoxylin and eosin, the nuclei of the epithelium of the tubules near the glomeruli, i. e., in the labyrinth, stain well while the cells of the straight terminal portions of the proximal convoluted tubules along the margins of the labyrinth are completely necrotic. In section *b*, stained by Gomori's method, the phosphatase in the epithelium within the labyrinth stains deeply and is in its normal position about the lumen. The phosphatase in the necrotic cells

is diminished in amount and is uniformly distributed throughout the necrotic mass. This dog's kidneys contained 11.06 units of phosphatase per gram of tissue.

PHOSPHATASE IN DOGS POISONED WITH URANYL NITRATE

The phosphatase of the blood and the kidneys of 15 dogs poisoned with uranyl nitrate was studied with the technic that was used in the study of the group poisoned with mercury bichloride. The effects are shown in table 5. One milligram or less of uranyl nitrate had little effect on blood phosphatase but increased renal phosphatase 30 per cent above the normal mean. On the other hand, doses of 2 mg. or more increased

TABLE 5.—*Determinations of Phosphatase in Normal Dogs and in Dogs Poisoned with Uranyl Nitrate*

	Average of Values in Bodansky Units			
	Before Poisoning	After Injection of 1 Mg. or Less of Uranyl Nitrate	Before Poisoning	After Injection of 2 Mg. or More of Uranyl Nitrate
Blood phosphatase	2.65 (?)	3.47	3.70	5.66
Renal phosphatase		14.09		16.29

blood phosphatase 53 per cent and renal phosphatase 52 per cent above the corresponding normal means.

Staining by Gomori's method revealed phosphatase diffusely spread throughout the cytoplasm of renal cells severely damaged by uranyl nitrate and phosphatase-containing detritus in the tubules, similar to that in mercury bichloride poisoning. With minimum necrotizing doses the distribution of these changes was also similar to that in the kidneys of dogs poisoned with mercury bichloride, i. e., the necrosis of the epithelium and the alteration in the enzyme were most marked in the distal portions of the proximal convoluted tubules. In regions of complete necrosis the phosphatase stained diffusely and sometimes deeply in the necrotic mass. Breedis, Flory and Furth [12] described this change as "phosphatase-rich casts."

Figures *2a* and *2b* are from the kidney of a dog (K Ur 22) that received two injections, on successive days, of 3 mg. of uranyl nitrate for each 100 cc. of blood and was put to death on the fourth day after the first injection. This amount was far above the minimum necrotizing dose. In figure *2a*, showing a section stained with hematoxylin and eosin, the epithelium of the tubules within the labyrinth is swollen and ragged but the nuclei stain, while the straight terminal portions along the margins of the labyrinth are necrotic. In figure *2b*, showing a section stained by Gomori's method, the phosphatase of the epithelium within the labyrinth stains well and is concentrated about the lumen but with slight diffusion through the cytoplasm, while the straight portion is necrotic and stains poorly but uniformly. In the left upper quadrant, beginning just below the glomerulus in the top center, is a tubule cut longitudinally through the origin of the straight terminal segment. In the upper end of this tubule the phosphatase is concentrated about the lumen but is moderately diffused through the cytoplasm. In the next short segment, where the tubule curves downward, cell outlines are lost and the entire mass is stained deeply, showing a high content of phosphatase. In the terminal straight portion of this tubule the epithelium is completely necrotic and stains palely but somewhat irregularly. In the right lower corner is a glomerulus with phosphatase-containing debris in its capsular space. This debris was probably regurgitated into the capsular space from necrotic material in more distal parts of the tubule, since the phosphatase in the epithelium in the beginning of the tubule, seen leaving the glomerulus, is almost normal in its distribution within the cells. The kidneys of this dog contained 21.70 units of phosphatase per gram of tissue.

PHOSPHATASE IN DOGS POISONED WITH POTASSIUM DICHROMATE

Studies of phosphatase were made on 9 dogs poisoned with potassium dichromate with the following results: In 5 dogs receiving 2 mg. or less for each 100 cc. of blood the renal phosphatase averaged 15.85 units per gram (47.85 per cent above the normal mean). In each of the poisoned animals the renal phosphatase was higher than the normal mean, with a range of 11.18 to 18.86 units. Four dogs received 4 to 6 mg. of potassium dichromate for each 100 cc. of blood in two doses of 2 and 3 mg. each. In this group the renal phosphatase values averaged 7.41 units (44.66 per cent below the normal mean), with a range of 6.0 to 9.6 units.

In sections of kidneys with minimal necrosis stained for phosphatase by Gomori's method the phosphatase in the terminal straight portions of the proximal convoluted tubules was limited to the brush border of the cells as in the normal controls. In the upper parts of the tubules, particularly in the subcapsular zone, and throughout the entire length of the proximal convoluted tubules

12. Breedis, C.; Flory, C. M., and Furth, J.: Arch. Path. **36**:402, 1943.

in dogs poisoned with larger doses, cell outlines were lost and the coagulated mass was stained uniformly black or pale brown.

The sections in figures 3*a* and 3*b* are from the subcapsular zone of a kidney of a dog (K Cr 16) that received two injections, on successive days, of 3 mg. each of potassium dichromate for each 100 cc. of blood. In the first section, stained with hematoxylin and eosin, there is massive necrosis of the epithelium of the first parts of the proximal convoluted tubules. Since the amount given this dog was greatly in excess of the minimum necrotizing dose, the necrosis involved practically the entire tubule and was not limited to the first segment. In the second section, stained by Gomori's method, the necrotic material of most of the tubules stains uniformly black, which indicates that active phosphatase is still present. The kidneys of this dog contained only 6.65 units of phosphatase per gram of tissue.

COMMENT

The results of the experiments described in detail in the foregoing pages are summarized in table 6. In animals poisoned with small doses of mercury bichloride and potassium dichromate, i. e., 2 mg. or less for each 100 cc. of blood, the activity of renal phosphatase was increased on the average above the normal mean (1 : 1.54 and 1 : 1.45, respectively) to an extent that appears not to be merely the result of chance; larger doses (3 mg. or more) caused marked reduction (1 : 0.85 and 1 : 0.69, respectively). This reduction was greater with potassium dichromate than with mercury bichloride, but the doses of potassium dichromate in some dogs reached a total of 6 mg. per hundred cubic centimeters of blood. The mean values of renal phosphatase in both groups poisoned with uranyl nitrate, i. e., those receiving 2 mg. or less and those receiving 3 mg. or more per hundred cubic centimeters of blood, were much greater than the normal means (1 : 1.30 and 1 : 1.52, respectively).

TABLE 6.—*Summary of Determinations of Phosphatase*

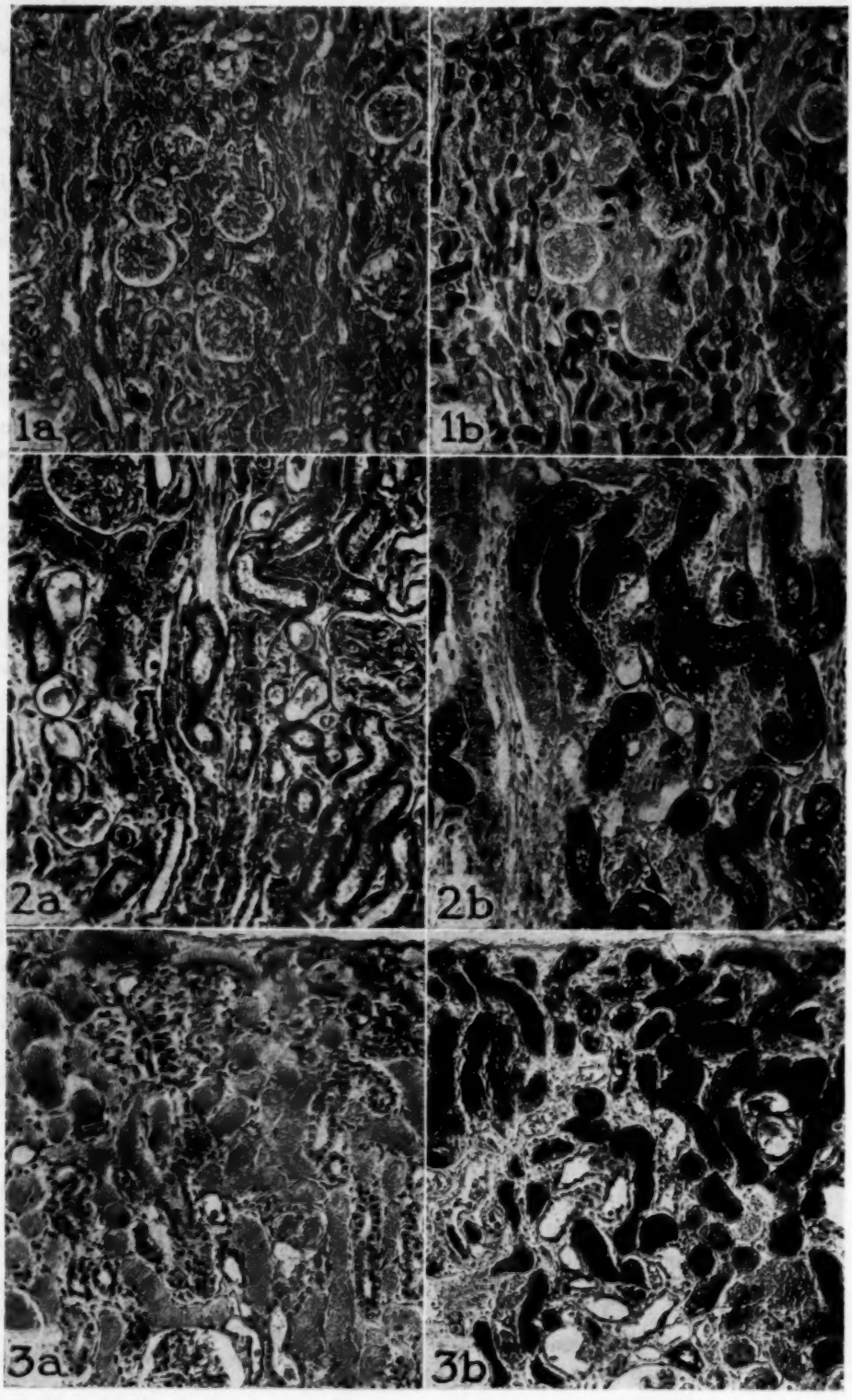
	Ratio Between Mean Values for Normal and Poisoned Dogs					
	$HgCl_2$		$UO_2(NO_3)_2$		$K_2Cr_2O_7$	
Phosphatase	2 Mg. or Less	3 Mg.	2 Mg. or Less	3 Mg.	2 Mg. or Less	3 Mg. or More
Blood.....	.....	.....	1:0.94	1:1	1:0.69	1:0.22 ?
Renal.....	1:1.54	1:0.85	1:1.30	1:1.52	1:1.45	1:0.69

Of 6 dogs given 3 mg. of mercury bichloride for each 100 cc. of blood, 5 died. One of those that died had been given only 2 mg. of this substance. None of the animals given uranyl nitrate or potassium dichromate in the doses stated died spontaneously within eight days, although the kidneys of those receiving the larger doses of both poisons showed extensive necrosis in their proximal convoluted tubules.

The activating and inhibiting effects of various substances on renal phosphatase have been studied. Erdtmann [13] discovered that alkaline phosphatase is activated by magnesium in proper concentration, and this has been confirmed by Kay [14] and others. Bamann,[15] Bamann and Heumüller,[16] D. Albers [17] and Massart and Vandendriessche [18] stated that this enzyme is activated by many kations, while Massart and Vandendriessche [18] and Miguel [19] observed that it is inhibited by anions. Thannhauser [20] and his coworkers reported that alkaline phosphatase is activated by iron, manganese, cobalt and nickel ions and is inhibited by zinc and copper. Perlman and Ferry [21] confirmed these observations on manganese and zinc.

Snow and Hepler [22] used multiple dilutions of a solution of 1 milligram-molecule $\times 10^{-3}$ to study the effects of the poisons used in these experiments on renal phosphatase. They observed that potassium dichromate and mercury bichloride caused definite but relatively slight reduction, while uranyl nitrate in the higher concentration employed induced marked reduction, in the activity of the enzyme. Magnesium chloride used as a control on the same material increased the activity of the enzyme by approximately 50 per cent. In view of these results it is difficult to interpret the consistent increase in the action of phosphatase in the kidneys of dogs poisoned with small doses of these substances. Three possibilities may be considered: Unknown factors in the living animal, not present in the test tube, may influence results. The enzyme may have been protected against the poisons by their combination with proteins of the cells in accordance with the well known phenomenon of precipitation of protein by heavy metals. Or,

13. Erdtmann, H.: Ztschr. f. physiol. Chem. **172**:182, 1927; **177**:211, 1928.
14. Kay, H. D.: J. Biol. Chem. **93**:733, 1931.
15. Bamann, E.: Naturwissenschaften **28**:142, 1940.
16. Bamann, E., and Heumüller, E.: Naturwissenschaften **28**:535, 1940.
17. Albers, D.: Ztschr. f. physiol. Chem. **266**:1, 1940.
18. Massart, L., and Vandendriessche: Naturwissenschaften **28**:143, 1940.
19. Miguel, E. J.: An. Asoc. quím. fram. Uruguay **42**:12, 1940.
20. Thannhauser, S. J.; Reichel, M., and Frattan, J. F.: J. Biol. Chem. **121**:697, 1937.
21. Perlman, G. E., and Ferry, R. M.: J. Biol. Chem. **142**:513, 1942.
22. Snow, A., and Hepler, O. E.: Personal communication to the authors.



Figures 1 to 3

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Explanations of Figures 1 to 3

Fig. 1 *a*.—Section of renal tissue from dog K Hg 6, which received one injection of 3 mg. of mercury bichloride to each 100 cc. of blood and died on the third day; hematoxylin and eosin stain; × 50. Nuclei of tubular epithelium within the labyrinth stain well; there is necrosis of the straight terminal portion of the proximal convoluted tubule along the margins of the labyrinth.

Fig. 1 *b*.—Serial section to the one shown in figure 1 *a*, but stained by Gomori's method; × 50. The phosphatase in the epithelium of the tubules within the labyrinth is practically normal in amount and distribution; it is markedly reduced in the necrotic epithelium. Chemically, the tissue contains 11.06 units of phosphatase per gram.

Fig. 2*a*.—Section of renal tissue from dog K Ur 22, which received two injections, on successive days, of 3 mg. of uranyl nitrate to each 100 cc. of blood and was killed on the fourth day; hematoxylin and eosin stain; × 155. There is marked necrosis of the straight portions of the proximal convoluted tubules; the nuclei within the labyrinth stain well, although the cells are not normal.

Fig. 2 *b*.—Section of renal tissue from the same dog as that shown in figure *2 a*, but stained by Gomori's method; × 155. There is abundance of active phosphatase with essentially normal intracellular distribution within the labyrinth; along the margins of the labyrinth the epithelium is necrotic and phosphatase activity is greatly reduced. In the left upper quadrant is a proximal convoluted tubule cut through the beginning of the terminal straight portion. In the upper end of this segment the phosphatase is approximately normal in amount and distribution; in the lower portion the epithelium is necrotic and phosphatase is only very slightly active; between these two portions the phosphatase is diffused through the cytoplasm but is not much reduced in activity. Chemically, the tissue contains 21.70 units of phosphatase per gram.

Fig. 3 *a*.—Section of renal tissue from dog K Cr 16, which received two injections of 3 mg. of potassium dichromate to each 100 cc. of blood on successive days and was killed on the fourth day; stained with hematoxylin and eosin; × 155. There is extensive necrosis of the epithelium of the proximal convoluted tubules in the subcapsular zone and throughout the full length of the tubules.

Fig. 3 *b*.—Section of renal tissue from the same dog as that shown in figure 3 *a*, but stained by Gomori's method; × 155. The phosphatase in the completely necrotic epithelium is active and gives a strong reaction. Chemically, the tissue contains 6.65 units of phosphatase per gram.

finally, the amount of the enzyme may be actually increased by the retention in the necrotic or damaged epithelium of phosphatase which has escaped from the blood in the glomerular filtrate.

If phosphatase in the kidneys has any function it would seem that this must, in some way, be bound up with its distinctive location in the brush border of the epithelium of the proximal convoluted tubules, where it is in immediate contact with the glomerular filtrate in the lumen. The phosphatase of the small intestine is in the superficial cells of the mucosa and is thus strategically located for taking part in the absorption of some substance, possibly dextrose, from the intestinal contents. In other organs, such as the liver (Gomori [2]; Kabat and Furth [23]) and the prostate (Huggins and Johnson [24]; Kutscher and Wolbergs [25]), this enzyme is distributed throughout the cytoplasm of the cells. Freeman, Chen and Ivy [5] found much phosphatase in the bile and believed that the enzyme "originated in the liver." Human semen, particularly the portion from the prostate gland (Huggins and Johnson [24]), contains acid phosphatase in high concentration (Kutscher and Wolbergs [25]; Guttmann and Guttmann [26]). Thus the acid phosphatase of semen is probably derived from the abundant supply in the cytoplasm of the cells of the prostate. These examples suggest that phosphatase which is uniformly distributed throughout the cytoplasm (liver and prostate) is either a product, or is concerned with the production of a substance, which is discharged into a lumen as an external secretion. On the other hand, the location of phosphatase in the brush border of the cells of the proximal convoluted tubules and in the superficial cells of the intestinal mucosa suggests that this enzyme in these organs is concerned with the absorption of some substance or substances from a lumen. Its alleged relation to absorption of dextrose from the glomerular filtrate and to deposition of calcium in the kidneys will be considered in subsequent papers in this series.

As this paper was being completed, a contribution from Russell, Rouse and Read [27] appeared. After referring to our previous paper [9] in which we stated that phosphatase could still be demonstrated by chemical and histochemical methods in cells rendered necrotic by salts of heavy metals, these authors continued: "This observation suggests an extrarenal origin for the alkaline phosphatase in the kidney since apparently the maintenance of the enzyme is not dependent on viable renal cells." It is not particularly surprising that an enzyme should remain active in dead cells since Moore, Goldstein and Cantowitz [28] have shown that marked histologic changes in the cytoplasm and the nuclei of renal epithelium may occur before the mitochondria—very intimate internal structures of the cell—show any pathologic alteration other than that of position, i. e., diffusion through the cytoplasm. Russell and his co-workers [29] found phosphatase in tubercles but "concluded that alkaline phosphatase does not appear in areas of caseation simultaneously with the development of necrosis of the cells." They stated that "there is some evidence to indicate that alkaline phosphatase in areas of caseation is not of autochthonous origin but is probably derived from the serum, the enzyme diffusing into the area of caseation." However, these authors [29] dismissed the idea of an extrarenal origin of phosphatase with the statement that "it seems most unlikely that the high concentration of alkaline phosphatase in the kidney could be totally explained by a special affinity of certain renal cells to pick up and store the enzyme that had been made in other cells of the body and transported to the kidney by the blood." While we did not make such a suggestion in our previous paper, we had formulated a concept of a possible extrarenal origin of renal phosphatase by an entirely different line of reasoning. The idea is not as fantastic as Russell and his co-workers imply by their comment.

Since foreign proteins with relatively large molecules, e. g., egg albumin, introduced into the blood stream are known to pass through the glomerular filter (Babcock [30] and others), it is likely that the alkaline phosphatase of the plasma may also escape through the glomeruli. It is present in the urine. The amount of a substance which the tubules are capable of absorbing is limited. There is also evidence that there is a limit to the size or the weight of the molecules which they can reabsorb from the glomerular filtrate. The largest molecule which the tubules regularly reabsorb is dextrose with a molecular weight of 180. The maximum capacity for reabsorption (the threshold value) of dextrose is about 180 mg. per hundred cubic centimeters,

23. Kabat, E. A., and Furth, J.: Am. J. Path. **17**: 303, 1941.

24. Huggins, C., and Johnson, A. A.: Am. J. Physiol. **103**:574, 1933.

25. Kutscher, W., and Wolbergs, H.: Ztschr. f. physiol. Chem. **236**:237, 1935.

26. Guttmann, A. B., and Guttmann, E. N.: Endocrinology **28**:115, 1941.

27. Russell, W. O.; Rouse, E. T., and Read, J. A.: Arch. Path. **38**:40, 1944.

28. Moore, R. A.; Goldstein, S., and Cantowitz, A.: Arch. Path. **8**:29, 1929.

29. Russell, W. O.; Read, J. A., and Rouse, E. T.: Arch. Path. **38**:31, 1944.

30. Babcock, C.: Anat. Rec. **71**:233, 1938.

or 0.001 mol. Stieglitz [31] injected a solution containing ferric ammonium citrate, which has a molecular weight of 724, into the veins of rabbits. He stated that this substance was excreted by the tubules. However, his illustration shows iron, rendered visible by the prussian blue reaction, sharply limited to the brush border and the immediately underlying very narrow zone as if it had penetrated the cells from the contents of the lumen. Inulin has a molecular weight of about 5,000 (Westfall and Landis [32]), but because of the great length of its molecule it has a diffusion coefficient equivalent to a molecular weight of about 15,000 (Bunim and co-workers [33]). Although inulin passes through the glomeruli readily, it is not reabsorbed by the tubules. Albers and Albers [34] have stated that the molecular weight of phosphatase is between 6,000 and 10,000.

Since substances of similar and even lower molecular weight are known not to be reabsorbed from the glomerular filtrate by the tubules it is possible that renal epithelium is unable to transfer such a large molecule as phosphatase through the cell substance into the surrounding tissue spaces. Stieglitz' [31] illustration strongly suggests the inability of the cells of the proximal convoluted tubules to transport molecules of ferric ammonium citrate which may have diffused through the brush border and cell membrane, through the remaining thickness of the cell even with the aid of the osmotic "pull" of the plasma proteins in the peritubal capillaries, augmented as it is by loss of water in the glomeruli. If a suitable method were available, it would be interesting to know whether the large molecules of inulin become entangled in the brush border and thus become concentrated in the same location where phosphatase is normally found. All this suggests the possibility that the presence of phosphatase in the brush border of the epithelium of the proximal convoluted tubules may represent an abortive attempt to reabsorb a substance of high molecular weight which has been excreted by glomerular filtration. If this admittedly bizarre and incongruous idea is correct, the presence of phosphatase in the kidneys may have no functional significance.

31. Stieglitz, E. J.: Am. J. Anat. **29**:33, 1921.

32. Westfall, B. B., and Landis, E. M.: J. Biol. Chem. **116**:727, 1936.

33. Bunim, J. J.; Smith, W. W., and Smith, H. W.: J. Biol. Chem. **118**:667, 1937.

34. Albers, H., and Albers, E.: Ztschr. f. physiol. Chem. **232**:165 and 189, 1935.

SUMMARY

All the experiments reported in this paper were made on dogs and the conclusions apply only to that species.

Changes above and below the normal mean occurred in the alkaline phosphatase of the blood serum within a period of eight days after poisoning with mercury bichloride.

Doses of potassium dichromate and of mercury bichloride that caused extensive necrosis of the epithelium of the renal tubules reduced the phosphatase activity of the kidneys. Uranyl nitrate in doses up to 3 mg. to 100 cc. of blood caused marked necrosis of the tubular epithelium but did not reduce the phosphatase activity of the kidneys as determined by chemical methods; in many animals the poison appeared to increase it.

Subnecrotizing doses of all three of the aforementioned chemical agents increased the activity of phosphatase in the kidneys, as determined by chemical methods.

The earliest morphologic change in the phosphatase in the epithelium of the proximal convoluted tubules induced by the heavy metal poisons used in these experiments was diffusion of the enzyme throughout the cytoplasm from its normal location in the brush border.

Active phosphatase can still be demonstrated by histochemical methods (Gomori's stain) in the debris of disintegrated necrotic cells of the proximal convoluted tubules.

The function of alkaline renal phosphatase is unknown. If it has a function, it would seem that this must be related to its limitation to the brush border of the epithelium of the proximal convoluted tubules and that it is concerned with absorption of some substance or substances from the contents of the lumen rather than the excretion of products into the lumen.

Reasons are presented for the possibility that alkaline renal phosphatase may not have any functional significance. Its presence in the brush border of the epithelium of the proximal convoluted tubules may be due only to an attempt by these cells to absorb it, the attempt being unsuccessful because its molecule is too large to be transported by the cells from the lumen to the peritubular tissue spaces.

ISCHIOPAGUS TRIPUS

REPORT OF TWO CASES

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An ischiopagus is an unusual type of symmetric diaxial monster in which fusion has taken place in the region of the perineum, with involvement of the bony pelvis. A secondary perineum consisting of tissue derived from both twins is present between the adjacent legs of each side, giving the impression of two pairs of legs and of genitalia arranged at right angles to the main axis of the body. This is the so-called cruciate monster or ischiopagus tetrapus. Actually, only one limb of each pair belongs to either of the individuals. If the main axes of the trunks meet at an angle, the two legs on the side of the smaller angle may fuse into a composite limb, resulting in the form known as ischiopagus tripus. These relations are indicated in the accompanying diagram (fig. 1).

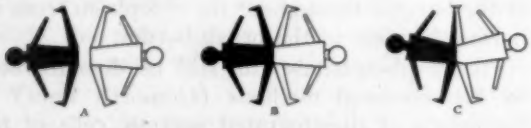

Fig. 1.—Diagram of the relationship of the components of an ischiopagus: *A*, two autonomous individuals. *B*, twins joined to form an ischiopagus tetrapus. *C*, twins with fusion resulting in an ischiopagus tripus. (The diagram is a modification of that published by H. H. Wilder in the American Journal of Anatomy **3**:387, 1904).

The earliest authentic case of this monstrosity was born in 1552 and reported two years later by Rueff.[1] In 1882 Taruffi[2] was able to find records of 14 undoubted cases of ischiopagus tripus; one which he overlooked was reported in this country by Ellis[3] in 1871. Most of these monsters lived only a few hours or days, none for as long as a year. The most recent account of the monstrosity is to be found in the Portuguese literature; it was described by Moitas[4] in 1941. The remarkable capacity of the fetus for functional adaptation in response to extensive developmental anomalies is well illustrated by the following 2 cases.

CASE 1

The mother was a healthy 24 year old white woman whose only previous pregnancy had resulted in a normal, full term infant. The father was apparently well. Among the mother's siblings was one set of twins, but there were none among the father's. The pregnancy was uneventful, and labor set in at the expected time. Six hours after the onset the head could be seen at the vaginal orifice. After birth of the head there was failure of restitution, and each arm was delivered manually. Further obstruction was then encountered, but by the use of traction and rotation the three legs and the body of the second twin were delivered.

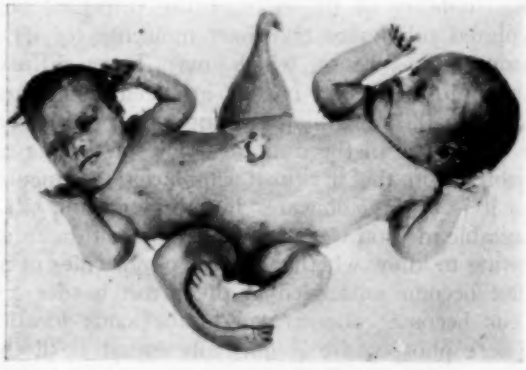

Fig. 2.—Photograph of the twins when 2 days old. The twin described as the smaller or right component is on the reader's left.

Immediately after birth the twins were cyanotic and motionless. Within a minute or two the larger one began to cry; the smaller one made the facial movements of crying but emitted no sound. The cyanosis disappeared rapidly except for that of the left half of the chest in the smaller twin, which persisted throughout life. The weight at this time was 11 pounds 12 ounces (5,330 Gm.).

The larger twin appeared normal to the level of the umbilicus. It breathed easily and was never cyanotic until shortly before death. The heart rate was 120 to 180 per minute; the sounds were normal in quality. Bottle-fed, it sucked well and cried lustily, although it often slept while the smaller twin was awake. It moved both arms freely, and when it kicked one leg, the composite third leg was also set in motion.

1. Rueff, J.: De conceptu et generatione hominis, Tiguri, C. Froschoverus, 1554, book 6, p. 47.

2. Taruffi, C.: Storia delli teratologia, Bologna, reg. tip., 1881-1891, vol. 2, pp. 366-403.

3. Ellis, C.: Boston M. & S. J. **8**:218, 1871.

4. Moitas, A.: Folia anat. univ. conimb. (art. 9) **16**: 1, 1941.

The smaller twin showed left-sided torticollis; no iris was present in the right eye, and in the left the pupil was irregular. Neither pupil reacted to light. No true respiratory excursions were ever observed, although some motion was transmitted by the diaphragmatic movements of the larger twin. When the latter cried, the former became cyanotic. The heart sounds were always muffled and too rapid to count. The sucking movements of this twin were poorly coordinated, and it took practically none of its formula. This member of the pair occasionally contorted its face as if to cry, but no sound was uttered. In general it was less active than the larger twin; yet it moved both arms freely,

Fig. 3.—The visceral relations as seen at autopsy. The cecum and the ascending colon pass transversely across the abdominal cavity. The umbilicus is to the left (the reader's right) of this structure. The umbilical vein can be seen passing to the liver of the left twin; the umbilical artery disappears in the pelvis. The left-sided position of the liver in the smaller twin is apparent, as is also the very large pericardial sac which completely fills the thoracic cavity.

and its leg synchronously with the composite one. At times it slept while the other was crying and threshing about. Urine and feces were expelled at normal intervals from the common urethral and anal orifices.

The twins gradually lost weight. Four days after birth the temperature of both rose to 102 F.; on the morning of the next day it reached 103 F. Respiration was rapid and the pulse rate 170 to 180. By evening the temperature was 105 F., the respiratory rate 80, and rales were heard at the bases of both lungs of the larger twin. Death occurred that night.

Autopsy.—The body weighed 8 pounds (3,629 Gm.); the over-all length from crown to crown was 47 cm. Rigor and livor mortis were present. The twin forming the right component of the monster was about a third smaller than that forming the left. A single umbilicus marked the area of fusion of the two trunks, which were apparently normal, as were also the upper extremities and the heads except for the abnormality of the eye in the smaller twin. At the level of the umbilicus and at right angles to the main axis of the body were two well formed legs, between which were normal female genitalia and an anus. Both feet showed talipes varus. In the same relative position on the opposite side was a fusiform structure that apparently had arisen from the union of two legs. The foot was represented by a sharply curved fleshy mass, 2.5 cm. long, with a single "toe nail" at its tip. At the base of this composite limb there were no vestiges of genitalia or an anus.

When the abdomen was opened, the distended large intestine was found to form a single loop arranged transversely to the main axis at the level of the umbilicus. Elsewhere in the larger twin the normal relations of the gastrointestinal tract, the liver, the pancreas and the spleen were preserved. However, in the right-sided member there was complete situs inversus of the corresponding organs, which were also much smaller than normal (fig. 3). Both small intestines opened into a common cecum, at the proximal end of which were two appendixes lying side by side. The cecum opened into a short large intestine, the lumen of which was double and emptied into a single rectum.

The lungs of the larger twin were pale pink, voluminous and covered the precordium. The heart was entirely normal. In the smaller twin the lungs were hidden by an enormous pericardial sac which completely filled the thoracic cavity and contained 70 cc. of clear yellow fluid. The lungs, deep purple, were wedged into the costovertebral angle and appeared to be quite airless. Histologically, the characteristic "crumpled paper bag" outline of the alveoli indicated that the lungs had never expanded—evidence that the pericardial effusion existed prior to birth.

The apex of the greatly enlarged heart was directed toward the right. It weighed 46 Gm. as compared with the normal heart of the larger twin, which weighed only 20 Gm. Dissection revealed transposition of the great vessels with an overriding aorta, a defect in the interventricular septum (fig. 4) and hypertrophy of the wall of the right ventricle. The aortic cusps were normal. The ascending aorta was a massive vessel which was continued cephalad as three large arteries—a right subclavian artery, a right common carotid artery and an innominate artery which arose to the left of these vessels. The arch of the aorta, directed to the right, was smaller than any one of these three arteries and looked more like a branch of the ascending trunk than a continuation of this vessel.

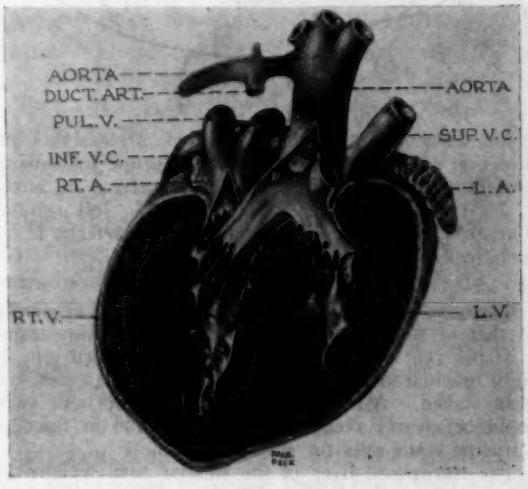

Fig. 4.—Drawing of the heart of the smaller twin. The section removed the anterior wall of each ventricle. The pulmonary artery which arose from the left ventricle, just anterior to the cut surface, is not shown. *Inf. V. C.*, inferior vena cava; *L. A.*, left auricular appendage; *L. V.*, left ventricle; *Pul. V.*, pulmonary vein; *Rt. A.*, right auricle; *Rt. V.*, right ventricle; *Sup. V. C.*, superior vena cava. *Duct. Art.*, ductus arteriosus.

The pulmonary artery arose from a stenotic opening in the left ventricle guarded by a malformed valve. It communicated with the aorta by a widely patent ductus arteriosus. The pulmonary veins combined to form a single large vein which emptied directly into

the right ventricle. The right auricle was represented by an inconspicuous and irregular little nodule attached to the wall of this vessel. The inferior vena cava joined the superior vena cava just before the latter emptied into the left ventricle. This junction was all that remained of the left auricle, although a fairly well developed auricular appendage was present. At the opening into the left ventricle was a well formed tricuspid valve. At the point of union between the superior and inferior venae cavae there was a valve leaflet so arranged that the blood flowing down the superior vena cava would pass into the inferior vena cava as well as into the left ventricle.

In the region of the pelvis the aorta of the smaller twin was directly continuous with the aorta of the larger by fusion with the right common iliac artery of the latter. It gave off a left common iliac artery which fused with the distal portion of the right common iliac artery of the larger twin to supply the single composite leg. From the left common iliac artery of the larger twin arose the single umbilical artery. Only a single, partly fibrosed umbilical vein was present, forming the ligamentum teres of the larger twin (fig. 3). Cross section of the cord likewise showed only one vein and one artery. From this it is apparent that in intrauterine life the placental circulation was directly associated only with the larger twin.

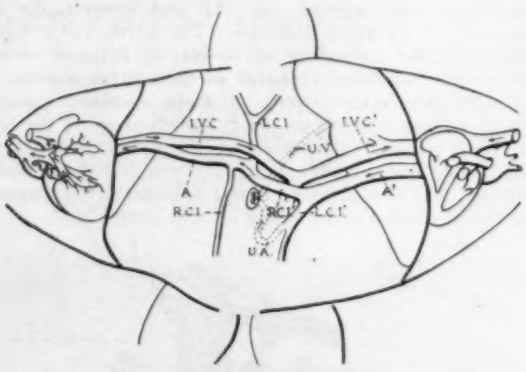

Fig. 5.—Diagram of the circulation; arrows show the direction of blood flow. The letters prime indicate the vessels in the left member of the monster. *A.*, aorta; *I. V. C.*, inferior vena cava; *L. C. I.*, left common iliac artery; *R. C. I.*, right common iliac artery; *U. A.*, umbilical artery; *U. V.*, umbilical vein.

The union of the inferior venae cavae was similar to that of the aortas. Before the autopsy was begun, a 35 per cent solution of diodrast was injected into the right brachial artery of the larger twin and a roentgenogram taken. All of the main vessels of both twins could be clearly recognized, as illustrated in figure 5, a tracing made over the roentgenogram.

The transposed liver of the smaller twin weighed 40 Gm.; that of the larger, 120 Gm. The gallbladders and biliary passages were apparently normal, as was also the histologic structure of the parenchyma of the livers. The spleens, which weighed 3 Gm. and 6 Gm. respectively, were grossly and microscopically normal except for the transposition of the smaller one.

A double kidney formed by end to end union was present in the pelvis at the level of the composite limb, its long axis parallel to that of the main axis of the two trunks (fig. 6). On section of the kidney the usual corticomedullary relations were present. However, scattered through the parenchyma were numerous discrete white areas (0.5 to 1 mm.) which on histologic examination were found to be abscesses. An infiltrate of neutrophils was likewise found in the pelvic mucosa.

The half of the double kidney belonging to the smaller twin possessed two ureters that fused to form a greatly dilated single ureter which opened into the bladder posteriorly. This saclike ureter may well have acted as an accessory bladder. No stenosis of the orifices was present. The remaining half of the kidney had a single ureter which passed behind the sigmoid flexure to open into the bladder anteriorly. Its distal portion for a length of 0.7 cm. was greatly constricted, yet the lumen was patent. Two urethral orifices were present at the base of the bladder. The urethras soon fused to form a single duct which opened externally in normal relation to the vagina and the rectum.

At each pole of the kidney was a roughly triangular adrenal gland. Two other adrenal glands, circular in outline, were found in their normal positions on the opposite side; their shape was due to the absence of the kidneys on that side.

Two uteri as well as four fallopian tubes and four ovaries were present in the pelvis (fig. 6). The tubes

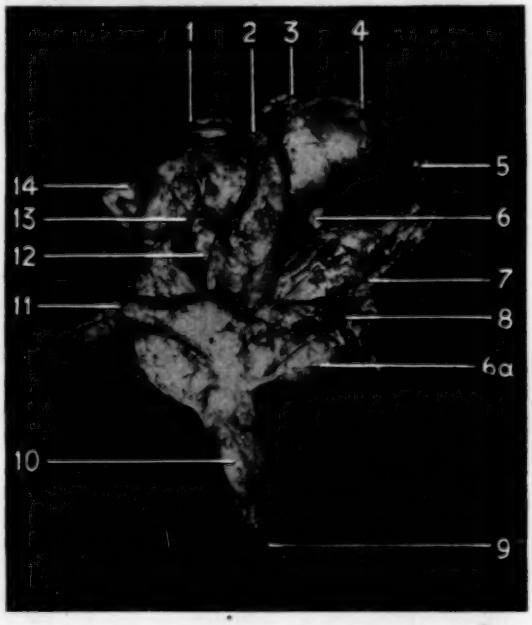

Fig. 6.—Photograph of pelvic viscera in situ; the adjacent structures have been blocked out. *1, 2, 3,* one set of fallopian tubes, ovaries and uterus; *4,* kidney; *5, 14,* adrenal glands; *6, 12, 13,* ureters; *7,* rectum; *8, 11,* second set of fallopian tubes, ovaries and bicornate uterus; *9,* urachus; *10,* bladder.

and ovaries were grossly and microscopically normal. The uterus on the side of the composite limb, though derived in part from each of the twins, was well formed and placed at right angles to the main axis of the body. The vagina lay between one of the ureters and the rectosigmoid; it had no orifice and was greatly distended with inspissated mucus. Fusion was incomplete in the second uterus, producing an extreme bicornuate structure in which each half lay parallel to the long axis of the trunk. The twin cervical canals opened into a single vagina, the orifice of which was normally placed between that of the urethra and the anus.

Except for a deformity of the right clavicle of the larger twin the bones of the trunks, the arms and the two legs were normal. The two lumbar vertebrae of each trunk were acutely flexed and laterally rotated. The central epiphyses of the single sacrum were found on the side of the two legs and at right angles to the

rest of the vertebral column. The lateral sacral epiphyses formed an arc that extended from the last lumbar vertebrae to a position lateral to the central epiphyses. The iliac, ischial and pubic bones were normally situated in relation to the sacrum (fig. 7).

On the side of the composite limb there was an irregular flat bone, apparently an ilium. The single femur had a poorly developed head and a shaft that flared out broadly at its distal end, where two epiphyses could be recognized. This is the chief evidence that the composite limb actually resulted from the fusion of two leg anlages early in embryonic development. The proximal end of the tibia was broad; the distal end, narrow. No fibula was present. A single bone could be recognized in the portion of the limb which represented the foot.

The brain of the smaller twin weighed 179 Gm.; that of the larger, 375 Gm. In both brains the gross structures and convolutional markings were normal, as were also the circles of Willis. In the brain of the smaller twin the veins were not collected into three vessels, but instead emptied directly into the longitudinal sinus. They were all greatly engorged and were present in such large numbers as to give the appearance of a veritable brush. On histologic examination of the brain substance the striking abnormality was a tremendous dilation of the capillary venous bed. Although these vessels were normal in arrangement, their size and their degree of engorgement gave them a prominence which at first led to the belief that there was also an increase in number. This, however, was more apparent than real.

Fig. 7.—Roentgenogram showing the bones of the trunk and the composite limb.

Fig. 8.—Gross appearance of the ischiopagus tripus in case 2. For a description see the text.

CASE 2

In the teratologic collection of the Army Medical Museum there is another example of ischiopagus tripus (fig. 8). Born of Filipino parents these twins survived for forty-six hours after birth; no further details of their history are available. The teras, which is a female, measured 36 cm. from crown to crown. When placed in a position similar to that employed in describing the previous specimen, the twin on the right was again distinctly smaller than that on the left. Likewise, it displayed developmental anomalies absent in the larger twin. There were a cleft palate and lip and a deformed right hand wherein only the thumb was separate, the fingers being united to form a mitten-like structure. The composite limb had two partly fused but recognizable feet, each foot bearing five toes.

On dissection the twin on the right exhibited complete situs inversus, which included both the heart and the abdominal viscera. The heart had a single ventricle and auricle; the lungs were completely collapsed. The aorta was broadly continuous with the right common iliac artery of the larger twin. The venae cavae were likewise confluent. The single umbilical artery arose from the left common iliac artery of this twin, which also received the only umbilical vein that could be recognized. Hence, it is apparent that the circulation was identical with that of the foregoing teras. All abdominal viscera were much smaller in the twin on the right than in that on the left. Fusion of the small intestines took place 6 cm. proximal to the cecum. The appendix, the large intestine and the rectum were single. A horseshoe kidney was found in the smaller twin; two normally placed kidneys were present in the larger. An adrenal gland surmounted the upper pole of each kidney, including the two components of the horseshoe kidney. The ureters opened into a single bladder that was transverse to the main axis of the trunks. A bicornate uterus with attached fallopian tubes and two ovaries was in normal relation to the bladder. No trace of a second uterus or adnexa could be found.

COMMENT

The ischiopagus belongs to the large group of terata known as symmetric diaxial conjoined twins. These consist of two fairly equally developed individuals that are partly fused. This union may occur in the region of the head (cephalopagus), the thorax (thoracopagus), the umbilicus (omphalopagus), the buttock (pygopagus) or the perineum (ischiopagus). Numerous subdivisions and interstages of these forms have been described. Both members are always of the same sex, which favors the view that they are derived from a single ovum, as are homozygous (identical) twins. This assumption also gains support from the fact that junction is always effected by a union of like parts; e. g., head to head, never head to buttock. The absence of haphazard fusion indicates that in every instance the developing embryos must have had parallel axes. This can be accounted for only by assuming a duplication of the developing ovum by fission some time between the two cell stage and the formation of the primitive streak. The subsequent union of the two individuals probably occurs during the first three weeks of development.

Considerable experimental evidence has been accumulated in support of these theories. In 1915 Huber [5] opened the uteri of rats at varying

5. Huber, G. C.: J. Morphol. **26**:247, 1915.

intervals after copulation. In 1 instance he found the two blastomeres resulting from the first cleavage distinctly separated by a space equal to about half the diameter of each of the cells. They were normal in size, shape and structure, as also were their nuclei. The two cells lay free in a slightly distended portion of the lumen and did not appear to have been separated as a consequence of manipulation. The authors suggested that each might have developed into a complete fetus, with production of identical twins. This possibility has since been realized by the work of Nicholas and Hall.[6] These investigators removed ova that were in the two cell stage from rat uteri and placed them in a slightly acid solution of sodium and potassium chlorides. The zona pellucida dissolved off in about three hours, and the two blastomeres could be readily separated by a jet of water. When returned to the uterus each underwent normal development. These authors also demonstrated the possibility of further development after fusion of two ova. Since they used unsegmented eggs rather than young embryos, they obtained a single giant fetus rather than a conjoined monster.

The production of double monsters by fusion of embryos in the gastrula stage was first carried out by Spemann in 1918, using the eggs of the salamander Triton. This material was reexamined and supplemented by Wessel,[7] who showed that when the blastopores of the two ova were placed in varying relations to each other, the type of double monster that would result from each relationship could be accurately predicted. The well known two-headed salamander first produced by Spemann in 1903 by constricting the egg with a hair loop is not relevant to this discussion since it is the result of a partial duplication of a single axis rather than of the fusion of two.

In the cases reported here there is complete situs inversus of the right component. As long ago as 1865 Förster[8] emphasized the frequency of situs inversus in one member of a double monster and stated that this occurred only in the twin on the right. Although there are exceptions to the rule limiting situs inversus to the individual on the right of a double monster, it is true for about 90 per cent of the cases. This interesting localization of the visceral anomaly was lost sight of by investigators until 1919, when Morrill[9] described it in three cruciate monsters of the fish Fundulus and a human dicephalus tribrachius. In the same year appeared a paper by Spemann and Falkenberg[10] on situs inversus in twins and double monsters. The authors provided a satisfactory explanation of this phenomenon based on their findings in identical twins and monsters experimentally produced in the salamander. The median limbs and trunk muscles of twins resulting from the division of a Triton embryo are poorly developed, whether this division occurs in the late gastrula or even in the two blastomere stage. Hence the twins, though separate, can be recognized as right and left. Of 25 "left" twins, 1 showed inversion of the heart; of 30 "right" twins, 14 had complete situs inversus. Of 12 double monsters with anterior duplication, the left portion was normal in all, and the right showed situs inversus in 10. The impaired development of the trunk musculature of the adjacent sides of the two twins resulted in a bending of the trunk away from the midline. In the left twin this was toward the left; in the right it was directed to the right. In both, subsequent development restored the normal position of the trunk. The first evidence of normal asymmetry is found in a twisting of the liver anlage to the right associated with a bending of the gut to the left. In the case of the left component of twins or double monsters, the abnormal curvature of the trunk to the left merely accentuates this normal asymmetry. However, in the right component the normal displacement of the gut to the left is counterbalanced by the bending of the trunk as a whole to the right. If this is sufficiently marked, the gut will be pulled to the right, and the liver anlage will take up a position to the left of it, leading to situs inversus viscerum.

This explanation of situs inversus does not apply to the same condition when found in an autonomous individual. In view of the prevalence of the anomaly in conjoined twins it was at one time suggested that when the anomaly was found in an otherwise normal person one could assume that the person was one of twins, the other having died early in development and been resorbed. This theory has been discredited, and Cockayne[11] has brought forth evidence to show that in these cases the tendency is inherited as a recessive characteristic.

Both the specimens of ischiopagus tripus here reported were female. Of the 14 recorded by Taruffi,[2] 11 were female; the sex of the remaining 3 is unknown. The one recently described by Moitas[4] may have been male. This preponderance of the female sex is found in most types of double monsters. It can probably be explained

6. Nicholas, J. S., and Hall, B. V.: J. Exper. Zool. **90**:441, 1942.

7. Wessel, E.: Arch. f. Entwcklngsmechn. d. Organ. **107**:481, 1926.

8. Förster, A.: Die Missbildungen des Menschen systematisch dargestellt, ed. 2, Jena, F. Mauke, 1865, p. 136.

9. Morrill, C. V.: Anat. Rec. **16**:265, 1919.

10. Spemann, H., and Falkenberg, H.: Arch. f. Entwcklngsmechn. d. Organ. **45**:371, 1919.

11. Cockayne, E. A.: Quart. J. Med. **7**:479, 1938.

in part by the fact that male fetuses are less likely to survive in the presence of developmental anomalies than are females. Potter and Adair [12] stated that stillborn fetuses delivered early in pregnancy are much more commonly male than female. Under four months 78 per cent are male. Many of the male monstrosities are therefore aborted at such an early date that anomalies are not readily recognized and are discarded by the physician. Furthermore, in the cases of miscarriage of early pregnancy the physician frequently does not even see the fetus, as he is called in only when persistent bleeding follows incomplete abortion.

The circulatory systems of these terata were remarkable for the broad union of the aortas via the right common iliac artery of the larger twin. Of added interest is the fact that in the case reported by Moitas a precisely similar union had occurred, indicating that given a certain set of conditions the same result will follow, whether in normal or in abnormal development. In view of the complete atelectasis of the lungs in the smaller twin, it was only the union of the aortas that made survival of the monster possible. Without it the intrauterine development would likewise soon have ended since the single umbilical vein and artery were in direct communication only with the blood supply of the larger twin. Although the direction of the flow of blood in the aorta and the inferior vena cava of the smaller twin was reversed, the blood in the aorta was arterial and that in the vena cava venous. If it is true that the blood flowed toward the heart in the descending aorta, then all its blood as well as that from the ascending aorta must have passed to the arms and the head. That this actually took place is indicated by the large caliber and engorgement of the cerebral vessels of the smaller twin in one of the monsters.

The heart of the larger twin was normal; that of the smaller, distinctly abnormal. Besides the transposition of the great vessels in the first case there were overriding of the aorta, a defect of the interventricular septum, stenosis of the pulmonary orifice and hypertrophy of the wall of the right ventricle. These findings can be classified together as forming the tetralogy of Fallot. Cockayne [11] pointed out that the most common cardiac anomaly in cases of complete situs inversus in an autonomous individual is, in fact, the tetralogy of Fallot. The author stated further that cardiac anomalies are present in almost 10 per cent of cases showing transposition of the viscera.

12. Potter, E. L., and Adair, F. L.: Fetal and Neonatal Death, Chicago, University of Chicago Press, 1940, p. 4.

The great pericardial effusion surrounding the anomalous heart of the first monster was not associated with any inflammatory reaction. However, in the epicardium were very many greatly distended thin-walled vascular channels. Comparison with the same region in the normal heart of the larger twin showed that these vessels were five times as numerous in the abnormal heart and their calibers many times greater. The dilation of these veins and the increased permeability of their walls due to the anoxemia of this twin before and after birth may account for the large transudate present in the pericardial sac.

The presence of a fused lateral kidney in the first case and of a median horseshoe kidney in the second fits in well with the modern concept of the formation of compound kidneys as described by Lewis and Papez.[13] These investigators, basing their interpretation on the study of a large series of pig embyros, stated that the renal anomaly was due to the relation of the kidneys to the aorta where the latter divides into the common iliac arteries. This bifurcation forms a crotch in which the kidneys are lodged and from which they escape by migrating upward. The arteries, therefore, act as a mechanical obstruction which tends to bring the renal blastemas close together, so that fusion may readily take place. A precisely similar condition was present in the first case here described. The right renal blastema of one individual and the left of the other were caught in the crotch of the partly fused common iliac arteries that supplied the composite limb. Hence a lateral union resulted after the manner of the more common median fusion in autonomous individuals as is seen in one member of the second ischiopagus here reported.

The incomplete double ureter of one component of the laterally fused kidney in case 1 may be accounted for by an exaggeration of the normal bifurcation of the tip of the ureteral stalk. Instead of being confined to the primitive pelvis the fission was continued into the ureter. We can offer no explanation for the unilateral renal agenesis.

SUMMARY

In 2 cases of ischiopagus tripus the right member of the pair suffered a complete situs inversus viscerum associated with extensive cardiac anomalies. Likewise, in each teras there was an end to end fusion of the aortas so that the flow of blood in the aorta of the member on the right was reversed.

13. Lewis, F. T., and Papez, J. W.: Anat. Rec. **9**: 105, 1915.

STUDIES ON THE PATHOGENESIS OF GLOMERULONEPHRITIS

I. PRODUCTION OF AUTOANTIBODIES TO KIDNEY IN EXPERIMENTAL ANIMALS

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In reviewing the literature which deals with the numerous attempts of many investigators along several distinct lines of experimentation to produce glomerulonephritis in animals, it becomes apparent that only those procedures in which specific antibodies to kidney were employed as the pathogenic agent have met with any notable and consistent success. Although renal lesions have been produced by the use of various micro-organisms and nontoxic antigens, such as foreign serum, these changes have not been shown to be progressive as is human glomerulonephritis in a considerable proportion of cases. However, the lesions obtained by means of antikidney serums have been shown [1] to be of a severe and frequently progressive nature and are regarded as resembling human glomerulonephritis in many respects much more than the changes produced by agents other than renal antibodies.

The possibility therefore suggested itself that human glomerulonephritis might be due to antibodies reacting specifically to renal material. Such an interpretation, however, seemed difficult, particularly as to the formation of such antibodies, since in the experiment the antikidney serum had to be prepared in an animal species other than the one in which the nephritis was to be produced.

If human glomerulonephritis is caused by specific antikidney antibodies, it would have to be assumed that the subject's own kidneys furnish the antigen for the production of these antibodies. Investigations have been undertaken wherein an attempt was made to produce antibodies in response to kidney of the same species by means of repeated injections of rabbit kidney, either fresh or autolyzed,[2] in rabbits; however, the results were entirely negative both with respect to the formation of renal antibodies reacting in vitro and with respect to the production of nephritis.

The first evidence in favor of the possibility of antigenic action of renal material of the same species under certain circumstances was brought forth by Schwentker and Comploier.[2b] They repeatedly injected mixtures of rabbit kidney emulsion and staphylococcus toxin into rabbits. By means of complement fixation, with plain rabbit kidney emulsion used as an antigen, they recorded positive reactions with the serum of the immunized animals up to a maximal dilution of serum of 1:80. But when the treatment consisted of injections of mixtures of rabbit kidney and streptococcus toxin (Dick toxin) they obtained only very faint reactions up to a serum dilution of 1:10. No reaction was detected with serum from rabbits which had been treated with either one of the toxins alone or with rabbit kidney alone.

The purpose in the present studies has been (1) to investigate more closely the possibility of a production of autoantibodies to kidney incited by homologous kidney rendered antigenic by combination with foreign antigens—particularly streptococci and their products—and (2) to ascertain whether any antibodies to kidney formed have the ability to react with the kidney in vivo, thus precipitating nephritis.

The studies have been carried out simultaneously in two animal species, rabbits and rats, by means of essentially analogous procedures.

MATERIALS AND METHODS

Preparation of Renal Material.—In order to prevent any extensive interference by the antigens of the blood, the kidneys which were to be used for immunization

From the George Williams Hooper Foundation for Medical Research and the Division of Medicine of the University of California Medical School.

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1. Wilson, G. W., and Oliver, J.: J. Exper. Med. **32**:183, 1920. Masugi, M.: Beitr. z. path. Anat. u. z. allg. Path. **91**:82, 1933; **92**:429, 1934. Smadel, J. E.: J. Exper. Med. **64**:921, 1936; **65**:541, 1937. Swift, H. F., and Smadel, J. E.: ibid. **65**:557, 1937. Smadel, J. E., and Farr, L. E.: ibid. **65**:527, 1937. Smadel, J. E., and Swift, H. F.: ibid. **74**:345, 1941. Ehrich, W. E.; Wolf, R. E., and Bartol, G. M.: ibid. **67**:769, 1938. Hemprich, R.: Ztschr. f. d. ges. exper. Med. **95**:304, 1935.

2. (*a*) Parks, A. E.; Ethridge, C. B., and Taussig, B. L.: Proc. Soc. Exper. Biol. & Med. **35**:418, 1936. (*b*) Schwentker, F. F., and Comploier, F. C.: J. Exper. Med. **70**:223, 1939.

and serologic work were perfused until they were free of blood. This was done while the animal was under ether anesthesia by inserting a cannula either into the aorta near the branching of the renal arteries or, more recently, into the left ventricle. An outlet was provided by opening the right auricle. Isotonic solution of sodium chloride at 38 to 40 C. and under pressure of about 120 cm. of saline solution was used. If the perfusion was begun while the animal was still alive, the kidneys always became very pale in a few seconds. Perfusion was continued for at least three to five minutes.

The kidneys were stripped of their capsules, weighed and then ground in a mortar with sand. Saline solution was added gradually to make a 20 per cent suspension by weight. Coarse particles were eliminated by short centrifugation. The whole procedure was carried out under strictly sterile conditions. Sterility tests of the emulsions were set up. The materials then were immediately frozen and kept at —76 C. in the carbon dioxide ice box.

Preparation of Streptococcic and Other Bacterial Antigens.—In order to prevent the introduction of antigens other than those of streptococcic origin, the organisms (group A beta hemolytic streptococci) were grown on a synthetic medium prepared according to the description by Bernheimer.[3] The medium consisted in the main of casein hydrolysate, additional amino acids (glutamine was found to be essential), salts, vitamins and dextrose. The acids which formed during the growth of the organisms were periodically neutralized. A heavy growth was obtained by this method. The organisms either were centrifuged out and resuspended in a smaller volume of saline solution or were concentrated in the medium by centrifugation. The supernatant medium was passed through a Seitz filter and used as a toxin. The bacterial suspensions that were used were from 2 to 7 per cent packed bacteria when centrifuged in Hopkins tubes. The bacterial suspensions usually were kept frozen at —76 C.

Staphylococcus strain "Wood" was grown on Bernheimer's synthetic medium for streptococci modified slightly to contain less dextrose (0.002 per cent) and, in addition, 0.08 per cent aminoacetic acid. The filtrate of these cultures in amounts of 0.3 cc. per kilogram of body weight killed rabbits in two hours.

Serologic Tests for Antibodies to Kidney.—The collodion method was used predominantly for the detection and the determination of antibodies to kidney. Other serologic tests occasionally were employed for confirmatory purposes. The collodion particle technic as used in these studies, especially with regard to the preparation of suitable particles (giving no nonspecific reaction), as well as to the optimal proportions of the components of the admixture, has been described in detail in another paper.[4] A brief outline of the test follows: As an antigen for the determination of antibodies to kidney, extracts of rabbit or rat kidney were used which had been prepared simply by centrifugation of the kidney emulsion until the supernatant was clear. When this was difficult to achieve, the supernatant was filtered through paper or a Seitz filter. These extracts (of 20 per cent kidney emulsions) had a protein content of approximately 0.2 per cent. Equal amounts of collodion stock suspension (of a density to match McFarland[5] scale no. 2 when diluted 1:20) and kidney extract were mixed immediately before use. The mixture was diluted 1:10 or 1:20 and added in amounts of 0.2 cc. to 0.5 cc. of the diluted serum, the latter having been prepared by the doubling dilution method in agglutination tubes, usually beginning with a dilution of 1:10. The tubes were shaken, incubated for one hour at room temperature and then centrifuged for three minutes at 1,400 revolutions per minute. The agglutinations were read while the sedimented particles were resuspended carefully by shaking.

Controls.—Each serum was tested in low dilutions (1:10 to 1:80) for nonspecific reactions with plain collodion particles. As soon as significant reactions of this sort were noted, new collodion particles were prepared. In addition, with each label of serum one or two normal serums were tested similarly with collodion antigen. Controls comprising extract of kidney plus collodion particles in saline solution also were set up. The titers were recorded in terms of serum dilutions with disregard for the further dilution which took place on the admixture of the antigen.

Satisfactory performance of the collodion method with tissue antigens had been investigated first with rat kidney and rabbit anti-rat kidney serum.

EXPERIMENTAL DATA

A total of 83 rabbits in groups comprising 3 to 10 animals were immunized with the following materials:

1. A strain of beta hemolytic streptococci isolated from a patient with sore throat, killed by heat (two hours at 60 C.) and mixed with rabbit kidney emulsion.
2. Ten separately grown strains of group A, beta hemolytic streptococci isolated from various persons with streptococcic infections, mixed, killed by heat and admixed with rabbit kidney emulsion.
3. The last-mentioned strains, mixed, killed with 1 per cent by volume of chloroform, admixed with rabbit kidney emulsion and incubated overnight at 37 C.
4. Strain N.Y. 5 of beta hemolytic streptococci, killed with 10 per cent by volume of ether and mixed with rabbit kidney.
5. Streptococcus strain N.Y. 5, living, mixed with kidney.
6. Dick toxin (filtrate of culture of strain N. Y. 5) plus rabbit kidney.
7. Streptococcus strain N.Y. 5, killed by ether and mixed with "kidney broth." The broth was prepared by heating the kidney emulsion in the autoclave at 20 pounds (9 Kg.) of pressure for three hours. The precipitate was removed entirely by passage through a Seitz filter. The filtrate was practically free of protein.
8. Streptococcus strain N.Y. 5 grown on rabbit kidney emulsion for twenty-four hours, with and without admixture of Bernheimer's synthetic medium to enhance growth, and killed by addition of ether. In some instances, the material was precipitated with alum instead of the admixture of ether.
9. Three other strains of group A streptococci grown on rabbit kidney emulsion and killed by ether.
10. A Seitz filtrate of rabbit kidney emulsion on which strain N.Y. 5 had been grown.
11. Staphylococcus toxin mixed with rabbit kidney.
12. Hog serum mixed with rabbit kidney.
13. Rabbit kidney emulsion.

3. Bernheimer, A. W.; Gillman, W.; Hottle, G. A., and Pappenheimer, A. M., Jr.: J. Bact. **43**:495, 1942.

4. Cavelti, P. A.: J. Immunol. **49**:365, 1944.

5. McFarland, J.: J. A. M. A. **49**:1178, 1907.

14. The same strains of beta hemolytic streptococci as used in combination with kidney alone, killed by heat, killed by ether, and living.

15. Staphylococcus toxin.

Proportions of the Components of the Admixtures.—Various proportions were used. The foreign antigen was admixed in amounts of about one tenth to one sixth of the amounts of renal material in terms of dry residue of each. After admixture the preparations, if not used immediately, were kept frozen at —76 C.

Schedules of Immunization.—In the earlier experiments injections were given twice weekly. With many rabbits these injections were continued until the animals had received more than forty injections. Schedules comprising three injections each week on successive days, a rest of one week being allowed after every two weeks, and schedules of five to ten injections on successive days, also were employed. Sometimes this treatment was repeated after a rest period of two to six weeks or more.

Mode of Injection and Dosage.—The injections were given intra-abdominally; 0.3 to 10 cc. of the antigen preparations (mixtures) were given each time, the maximal dose in most cases not exceeding 6 cc.

Bleeding.—The animals were bled from the ear vein on the fifth to eighth day after the last injection. In some cases frequent bleedings were carried out to determine the peak of antibody production.

Control Tests.—Before immunization the serum of each rabbit was tested for the presence of antibodies to kidney. In no case of untreated rabbits could any such antibodies be detected.

RESULTS OF IMMUNIZATIONS OF RABBITS

The serum of each rabbit was tested serologically after every two to six injections. None of the 17 control rabbits treated with streptococci, staphylococcus toxin or plain rabbit kidney showed any evidence of production of antibodies to kidney although some of them had been immunized for a long time and with considerable amounts of the antigen (up to fifty injections).

In all rabbits immunized with mixtures of a streptococcic antigen and rabbit kidney, antibodies to kidney developed, which were detectable by means of the collodion agglutination technic, an extract of plain normal rabbit kidney being used as an antigen for the test. The number of injections which were necessary varied considerably, but in several cases such antibodies were detected after two injections had been given, four days apart. No tests have been performed as yet after one injection. The maximum titer was reached after six to twenty injections; however, most of the animals reached their highest titer after twelve to eighteen injections. Subsequent, prolonged immunization usually resulted in a reduction of the titer, but with some exceptions. The height of the maximum titers varied from 1:80 to 1:40,960 in terms of serum dilutions. In a great majority of the cases it lay between 1:160 and 1:1,280.

As determined serologically, preparations containing whole streptococci killed by heat, ether or chloroform and mixed with kidney showed the highest activity in producing antibodies to kidney. The immunization with living streptococci mixed with kidney yielded lower titers. Considerably lower titers were also obtained with Dick toxin plus kidney and staphylococcus toxin plus kidney. Streptococci plus kidney broth produced good titers. Treatment with the preparations consisting of streptococci grown on rabbit kidney resulted in most instances in low titers. Hog serum plus kidney gave only doubtful results. (Table 1 gives a summary of some of the maximal titers observed in rabbits.)

TABLE 1.—*Titers of Renal Antibodies of Some Rabbit Serums in Terms of the Highest Dilution of Serum or Antigen Yielding a Positive Result*

Rabbit	Immunized with	Collodion Technic	Highest Dilution of Serum in Which Agglutination of Kidney Cells Occurred	Precipitin Test (Highest Dilution of Antigen Yielding Precipitation)
13	Rabbit kidney + killed streptococci	1,280	.....	80
12		640	.....	..
14		1,280	640	40
16		320	...	..
20		40,960	1,280	160
43		160	80	..
38		320	160	..
45		1,280	160	..
68		320	160	..
47		40	160	..
28	Dick toxin + rabbit kidney	160	...	..
33		160	...	..
36	Rabbit kidney broth + killed streptococci	1,280	...	..
39		320	...	..
2	Staphylotoxin + rabbit kidney	80	...	..
24		320	...	..
50		80	...	..
76	Hog serum + rabbit kidney	20	...	..
78		20	...	..
9	Rabbit kidney	0	0	0
10		0	...	..
17	Streptococci	0	0	0
26		10 (?)	...	..

OTHER SEROLOGIC TESTS FOR ANTIBODIES TO KIDNEY

A number of rabbit serums were tested for agglutination of suspensions of rabbit kidney cells.

Perfused rabbit kidney, which had been kept frozen, was ground and suspended in saline solution. The particles were washed two to three times by centrifugation. Coarse particles were eliminated by centrifugation for one minute at 1,500 revolutions per minute. When tested shortly after preparation, such suspensions, which consisted mostly of fragments of kidney cells,

gave good agglutinative results. The suspension was admixed in appropriate density to the serum dilutions. After incubation of about one hour at room temperature the agglutination tubes were centrifuged at 1,400 revolutions per minute for three minutes, and the agglutinations were read while the tubes were shaken.

In essence the results paralleled those found with the collodion method. Some differences in titer were observed, however; also, more tendency toward nonspecific agglutination was noted.

In a few cases, antigen dilution precipitin tests were carried out with extract of kidney as an antigen. With serums that were relatively high titered in terms of collodion agglutination precipitation was obtained; it was, however, usually of a low order, occurring with antigen dilutions not exceeding 1:40 to 1:160. As these dilutions refer to the extract prepared by centrifugation of 20 per cent kidney emulsion, the actual dilution in terms of renal substance would be 1:200 to 1:800.

A limited number of serums have been tested for species specificity with extracts of rat kidney. No reaction has been noted.

Approximately 40 serums have been compared by means of the collodion method with respect to their content of antibodies to kidney and antibodies to the streptococcus used for immunization. No consistent parallelism was found between the two types of antibodies; some serums of high titer of antibodies to kidney showed low titers of streptococcic antibodies, and vice versa.

One high-titered serum obtained by immunization with mixtures of rabbit kidney and streptococci was absorbed with rabbit kidney broth. After absorption this serum gave no serologic reaction with broth of rabbit kidney but still reacted strongly with extract of rabbit kidney.

A few rabbits which showed antibodies to kidney in the serum were killed and their serum was tested with their own kidney as an antigen. The same or a stronger reaction than that with kidney of normal rabbits was noted.

TABLE 2.—*Absorption Experiments: Titers of Renal Antibodies in Terms of the Highest Dilution of Serum Yielding Agglutination*

	Unabsorbed					Absorbed with						
						Liver		Serum				
Serum	Kidney	Liver	Brain	Red Blood Cells	Serum	Liver	Kidney	Serum	Kidney	Liver	Brain	Red Blood Cells
1.....	320	160	..	..	40	0	320	0	320	..	..	..
2.....	160	80	..	..	20	0	160	0	80	..	..	..
3.....	320	80	..	..	..	0	320	..	...	..	..	..
4.....	160	40	20	40	160	..	...	0*	160*	40*	0*	0*
5.....	320	40	20	40	160	..	...	0*	160*	40*	0*	0*
6.....	320	80	20	40	80	0	320	0*	320*	80*	0*	0*

* The serums were absorbed with serum and red blood cells.

SPECIFICITY OF THE ANTIBODIES TO KIDNEY DEVELOPED IN RABBITS

Some serums of moderate and higher titers were tested serologically by means of the collodion technic with respect to organ specificity with antigens such as serum, red blood cells and brain of the rabbit. In all cases the reactions with these antigens were considerably weaker than those with kidney as an antigen. Frequently no reaction at all was noted with serum or brain. The reactions with liver usually were more pronounced.

A few absorption experiments were attempted with rabbit liver and rabbit serum, the results of which are given in table 2. From this table it is evident that the antibodies reacting with tissues other than kidney could be removed by absorption with these tissues without appreciably impairing the titers of the antibodies reacting with kidney.

PRODUCTION OF ANTIBODIES TO KIDNEY IN RATS

Approximately 60 rats were immunized intraperitoneally with mixtures of perfused rat kidney and beta hemolytic streptococci which had been killed by heat or ether; the same strains of organisms, proportions of components of the admixtures and schedules of immunization as with rabbits were used. The doses varied between 0.1 and 1.0 cc. Blood was taken by syringe from the tail vein.

About 50 per cent of the immunized rats showed development of antibodies to kidney when tested serologically with collodion particles sensitized with extract of rat kidney. The titers lay between 1:20 and 1:1,280. It was observed on repeated bleeding after immunization that in rats the appearance of antibodies to kidney varied over a relatively long period, from about four days to more than three weeks after the last injection. A considerable proportion of the animals therefore may have had such antibodies in the blood at another time than when tested.

As in the case of rabbits, it was found in an additional large number of rats that preparations consisting of streptococci grown on rat kidney had only slight anti-

genicity with respect to the formation of antibodies to kidney.

Control rats treated with plain rat kidney failed to show evidence of the production of antibodies to kidney, whereas controls treated with killed or living streptococci occasionally showed some, though very slight, serologic reaction with rat kidney.

COMMENT

The results presented provide evidence that streptococci are able in some way to confer antigenicity on renal material which ordinarily is not antigenic in the same species. The underlying mechanism might most easily be explained by the conception of haptens (represented in the kidney) being attached to a "carrier" of protein nature (streptococcus or some of its substances). Such combinations when injected lead to the formation of antibodies—not only to the streptococcus but also, and independently from the latter, to kidney of the same species. Such antibodies to normal kidney of the same species have been demonstrated in the present experiment by several serologic methods: agglutination of collodion particles sensitized with extract of kidney, agglutination of suspensions of renal cells and cell fragments, and in a few instances also by precipitation of extract of kidney by the serum. The antibodies determined by the collodion method also were found to react serologically with kidney from the same animal which furnished the serum.

The conception of the hapten mechanism is supported by the observation that other antigens, such as staphylococcus toxin, also can act as a carrier for the kidney material, rendering it antigenic. It seems likely that haptens of nonprotein character were involved in the experiments which consisted of the immunization of rabbits with rabbit kidney broth plus streptococci. However, as shown by the absorption experiment mentioned on page 151 renal materials which are not heat stable and which presumably are of protein nature or are nonprotein substances attached to protein also are rendered highly antigenic by the addition of streptococci.

Some results of the present studies seem to indicate that with the immunizing procedures employed two, and possibly more, different antibodies reacting with kidney in vitro are formed.

The renal lesions obtained in rabbits and particularly in rats are to be described in a later paper.

With respect to the genesis of glomerulonephritis in man, as already suggested by Schwentker and Comploier,[2b] streptococci or their substances could easily be conceived as the cause of a slight toxic damage of the kidneys during the height of a streptococcic infection, perhaps demonstrated clinically by the albuminuria and other urinary symptoms which are noted so frequently in cases of streptococcic infection. Undoubtedly, material from such damaged renal tissue could enter the blood stream during the process of reparation, perhaps still attached to the toxin which had acted on it. On reaching the sites of the formation of antibodies this complex would act as an antigen. Antibodies to kidney would be formed, which, if they reach a sufficiently high level, would precipitate glomerulonephritis by their specific reaction with the hapten represented in the kidney.

SUMMARY

Evidence is presented by means of experiments in rabbits and rats that group A beta hemolytic streptococci are able in some way to render renal material antigenic in the same species. The antibodies to kidney produced by immunization with combinations of streptococci and homologous kidney were demonstrated by several serologic methods in vitro.

CYSTS AND CYSTIC TUMORS OF THE MEDIASTINUM

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The mediastinum is the site of a large variety of cysts and cystic tumors, and the following classification is offered:

Type of Tumor	Derivation
I. Congenital	Ectoderm
1. Epidermoid cyst	Ectoderm and mesoderm
2. Dermoid cyst	Ectoderm, entoderm and mesoderm
3. Teratoma	
4. Pericardial celomic cyst	Mesoderm
5. Bronchial cyst	Entoderm and mesoderm
6. Esophageal cyst	Entoderm and mesoderm
7. Gastroenteric cyst	
(*a*) Gastric	Entoderm and mesoderm
(*b*) Enteric	Entoderm and mesoderm
8. Cystic lymphangioma	Mesoderm
II. Acquired	
1. Parasitic cyst, caused by Taenia echinococcus	
2. Neoplastic cyst, due to degeneration of a solid tumor	
3. Cystic hematoma, resulting from degeneration of hematoma	

Of considerable interest are those thought to be congenital. In this paper the cases of 3 patients with such mediastinal tumors are reported; the genesis of the various types is discussed, and a review of the literature is presented.

EPIDERMOID CYST, DERMOID CYST AND TERATOMA

The epidermoid cyst, the dermoid cyst and the teratoma make up the largest group of intrathoracic congenital cysts and tumors. Although not rare, they are uncommon, as indicated by Hare's [1] review. Of 288 mediastinal tumors (including metastatic growths) he found only 11 to be of this congenital type.

In 1825 Gordon [2] reported the first authentic case of congenital mediastinal dermoid cyst, containing hair, sebaceous material, a rudimentary mandible and six teeth. A little more than one hundred years later, in 1933, Hedblom [3] collected reports of 185 cases of epidermoid cyst, dermoid cyst and teratoma from the literature and recorded 6 cases of his own, making a total of 191. A review of the subsequent literature up to January 1944 reveals 52 additional verified cases.[4] To these are added a subsequently reported instance of cancerous teratoma [5] and case 1 of this

From the Institute of Pathology, Western Reserve University and the University Hospitals of Cleveland.

1. Hare, H. A.: The Pathology, Clinical History and Diagnosis of Affections of the Mediastinum Other Than Those of the Heart and Aorta, Philadelphia, P. Blakiston's Son & Co., 1889; cited by Houghton.[41]

2. Gordon, J. A.: Med.-Chir. Tr., London **13**:12, 1827; cited by Hedblom.[3]

3. Hedblom, C. A.: J. Thoracic Surg. **3**:22, 1933.

4. (*a*) Harrington, S. W.: J. Thoracic Surg. **3**:50, 1933; **7**:191, 1937. (*b*) Pasman, R. E., and Pepe, C. J.: Rev. Asoc. méd. argent. **47**:2992, 1933. (*c*) Lambret, O.: Echo méd. du Nord **37**:445, 1933. (*d*) Bevan, A. D.: S. Clin. North America **13**:1165, 1933. (*e*) Hablützel, C.: Schweiz. med. Wchnschr. **63**:1308, 1933. (*f*) Kantrowitz, A. R.: Am. J. Path. **10**:531, 1934. (*g*) Hammarskjöld, B.: Acta radiol. **15**:210, 1934. (*h*) Divis, J.: Časop. lék. česk. **74**:18, 1934. (*i*) de Castro, J. R., and Parreira, H.: An. de med. int. **3**:935, 1934. (*j*) Reddingius, T.: Nederl. tijdschr. v. geneesk. **78**:5334, 1934. (*k*) Cisneros, R.: Bol. y trab., Soc. de cir. de Buenos Aires **19**:452, 1935. (*l*) Houghton, J. D.: Am. J. Path. **12**:349, 1936. (*m*) Piéchaud, F.; Poinot, J., and Bordenave, J.: Bull. et mém. Soc. méd. et chir. de Bordeaux, 1936, p. 56. (*n*) Moir, P. J.: Brit. M. J. **1**:463, 1936. (*o*) Somolinos, G.: Arch. cardiol. y hemat. **17**:1752, 1936. (*p*) Stanbury, W. S., and Oille, W. A.: J. Tech. Methods **16**: 52, 1936. (*q*) Schenck, S. B.: M. Rec. **144**:276, 1936. (*r*) Jellen, J., and Fischer, W. E.: Am. J. Dis. Child. **51**:1397, 1936. (*s*) Fox, J. P., and Hospers, C. A.: Am. J. Cancer **28**:273, 1936. (*t*) Leuret, E.; Piéchaud, F., and Delachaud, P.: Rev. de méd., Paris **53**:61, 1936. (*u*) Lanza, G.: Arch. per le sc. med. **63**:71, 1937. (*v*) Fanano, V.: Riv. di pat. e clin. d. tuberc. **11**:453, 1937. (*w*) Wheatley, G. M.: Am. J. Dis. Child. **54**:1057, 1937. (*x*) Smith, E. V., and Mills, R. G.: J. Thoracic Surg. **7**:338, 1938. (*y*) Rudenko, O. M.: Vrach. delo **20**:511, 1938. (*z*) Kuzma, W.: Polski przegl. radiol. **13**:145, 1938. (*a'*) Becker, B. J. P.: South African M. J. **13**:659, 1939. (*b'*) Doran, W. T., and Lester, C. W.: J. Thoracic Surg. **8**:309, 1939. (*c'*) Heuer, G. J., and Andrus, W. D.: Am. J. Surg. **50**:143, 1940. (*d'*) Gray, H. K., and Woodruff, R.: Minnesota Med. **23**:781, 1940. (*e'*) Aguilar, H. D.: Publ. d. centro de invest. tisiol. **4**:309, 1940. (*f'*) van Joost, C. R. N. F., and Kopp, J. G.: Geneesk. tijdschr. v. Nederl.-Indië **81**:969, 1941. (*g'*) Cabot Case 27052, New England J. Med. **224**:207, 1941. (*h'*) Amorim, A.: Bol. coll. brasil de cirurgiões **17**:17, 1942. (*i'*) Beck, C. S.: Ann. Surg. **116**:161, 1942. (*j'*) Friedrich, R.: Zentralbl. f. Chir. **69**:479, 1942. (*k'*) Carlson, H. A.: J. Thoracic Surg. **12**:376, 1943. (*l'*) Gebauer, P. W.: ibid. **12**:458, 1943. (*m'*) Sarroca, J. L.: Bol. Soc. cir. d. Uruguay **14**:52, 1943. (*n'*) Neuhof, H.: J. Mt. Sinai Hosp. **10**:402, 1943. (*o'*) Gravano, L.; Itoiz, O. A., and Bianchetti, S. L.: Rev. Asoc. méd. argent. **57**:878, 1943.

5. Laipply, T. C., and Shipley, R. A.: Extragenital Chorion-Epithelioma in the Male, Am. J. Path., to be published.

paper, making a total of 245 cases of epidermoid cyst, dermoid cyst and teratoma.

Origin.—Many hypotheses have been advanced to explain the origin of dermoid and teratoid tumors, but the exact derivation of the tissues of which they are composed has never been definitely determined. Two fundamentally different hypotheses are the monogerminal and the bigerminal.

The monogerminal hypothesis implies that in every case the tumor develops from one embryo. One of the proponents of this theory suggested origin by invagination of the ectoderm at the time of the closure of the primitive wall of the thorax. This concept, however, was based on the incorrect belief that these cystic tumors were made up only of tegmental structures. Other suggestions include origination from branchial clefts, thyroid gland and bronchi but fail to explain the complex tumors with three embryonal derivatives. Perhaps the most satisfactory hypothesis of the monogerminal type is the one indicating development from totipotent cells in embryonal rests of the urogenital fold.

The bigerminal hypothesis maintains that there is a second independent embryonal anlage which never reaches normal body form or structure, remaining as a parasitic fetus in fetu.

Some writers contend that the complex teratoma is of bigerminal origin, while the more simple dermoid and epidermoid cysts are explained by the monogerminal hypothesis.

Even though these theories offer some explanation for the development of the tumor of this group, they do not elucidate the fact that many of these tumors remain dormant or insignificant in size for years before they enlarge and produce symptoms.

Sex and Age of Patients.—Of the 245 patients whose cases are reported in the literature, 106 were males and 118 were females; in 21 cases the sex was not stated.

The incidence with respect to the age at the time when the diagnosis was established by operation or autopsy may be stated as shown in table 1.

TABLE 1.—*Age Incidence*

Age Group	Number	Percentage
Under 12 years	31	12.7
12-16 years	19	7.8
17-20 years	34	13.8
21-30 years	87	35.5
31-40 years	30	12.2
41-50 years	13	5.3
51-60 years	9	3.7
61-70 years	1	0.4
Age not stated	21	8.6

Note: For convenience the age groups are those used by Hedblom.[8]

The youngest patient was a stillborn infant; the oldest, a 62 year old woman. As indicated in the tabulation, these tumors are most common in the younger age groups, 69.8 per cent occurring in those less than 31 years of age.

Structures.—Hedblom's classification divides the congenital intrathoracic tumors into three groups: epidermoids, dermoids and teratomas.

The epidermoids, containing only derivatives of ectoderm, are cysts lined with stratified squamous epithelium. Their walls are made up of dense fibrous tissue with or without glands of ectodermal origin. They are filled with clear or milky fluid or with a gelatinous or pasty material, frequently mixed with hair.

The dermoids are also cysts and have tissues of mesodermal as well as ectodermal origin. Thus, in addition to hair, epithelium and glands, there are cartilage, bone, teeth, smooth or striated muscle and other structures.

The teratoma on microscopic examination shows tissues derived in part from entoderm, as well as from ectoderm and mesoderm. It is usually more solid than the other congenital tumors and contains varying combinations of tissues derived from the digestive tract and its associated glands and from the respiratory tract, the thyroid gland and the thymus.

The 245 cases in the epidermoid, dermoid and teratoma groups are classified as follows:

Epidermoid group
- Microscopically examined..... 48
- Not microscopically examined..... 59

Dermoid group
- Microscopically examined 47
- Not microscopically examined 21

Teratoma group
- Microscopically examined 59
- Not classified 11

Two hundred and seventeen (88.6 per cent) of the growths were benign, and 28 (11.4 per cent) were cancerous.

Symptoms and Signs.—The two most common symptoms were cough and pain in the chest. Cough, which was present in 115 patients, was productive of more or less purulent sputum in 49. Hemoptysis was noted in 35, and hair was coughed up by 33 of the patients. Pain in the chest with or without radiation to the shoulders or the arms was a prominent symptom in 78. Other symptoms included dyspnea in 24, marked loss of weight in 15, palpitation in 8, hoarseness in 8, dysphagia in 6 and weakness in 4.

There was dulness or flatness to percussion in 97, bulging of the chest in 38, cyanosis in 20, edema of the arms or the neck in 13, fever in 14,

displacement of the heart in 11, enlargement of cervical and thoracic veins in 12, signs of pleurisy with or without effusion in 24, signs of empyema in 10 and signs of pericarditis in 9.

As indicated by the age incidence, there was frequently a latent period of several years during which the tumor increased in size sufficiently to cause recognized symptoms, and in the majority of instances the onset of symptoms was insidious. The duration of the symptoms up to the time at which the diagnosis was established by operation or autopsy in 151 patients was less than six months in 45, six months to one year in 26, one to five years in 53, five to ten years in 21, ten to twenty years in 5 and thirty to forty years in 1.

Prognosis and Treatment.—Without treatment the prognosis varies with the size, the location and the rate of growth of the tumor. Some of the tumors remained small, produced only minor or no symptoms and were incidental unexpected observations at autopsy.

Roentgen treatment was of no value with the benign tumors and had no lasting effect in the cancerous ones. Death occurred in all patients with cancerous tumors so far as the result was stated.

The prognosis with surgical treatment also varies with the size and the location of the tumor. The first case in which a patient was operated on was that reported by Pöhn[6] in 1871; he drained a dermoid that presented in the neck. Complete extirpation of such a tumor was first successfully accomplished in 1893, by Bastianelli.[7] In recent years the development of thoracic surgery has led to successful surgical treatment in many cases.

The results of surgical treatment in the cases reported in the literature are indicated in table 2.

TABLE 2.—*Results of Surgical Treatment*

Treatment	Cases	Patients Cured	Patients Improved	Patients Not Improved	Patients Who Died	Cases in Which Result Is Not Stated
Drainage	22	0	11	1	8	2
Marsupialization and/or partial excision	28	10	15	0	3	0
Complete excision	76	55	7	0	12	2

Of the 126 patients subjected to surgical treatment, 65 (or 51.5 per cent) were cured, 33 (or 26.2 per cent) were improved, and 23 (or 18.3 per cent) died. The most common postoperative complication was a persistent bronchopleural fistula. Complete extirpation of the tumor cured 55 (or 72.4 per cent) of 76 patients so treated. Thus, the treatment of choice is radical excision. This is best accomplished before pressure symptoms or infection have supervened.

6. Pöhn, H.: Beschreibung eines Falles von Dermoidcyste des Mediastinum anticum, Inaug. Dissert., Berlin, G. Lange, 1871; cited by Hedblom.[8]

7. Bastianelli, R.: Riforma med., May 20, 1893; cited by Hedblom.[8]

PERICARDIAL CELOMIC CYSTS

The pericardial cysts are thought to result from an anomalous development of the pericardium, which is formed by the fusion of multiple disconnected lacunae. If one of the lacunar cavities failed to merge with the others, it could persist and develop into a pericardial celomic cyst. If, on the other hand, the rate of development of the primitive cavities was unequal, an unusually large lacunar space in continuity with the others could result in a congenital diverticulum of the pericardium.

The pericardial celomic cysts have thin, loose or dense fibrous connective tissue walls and are lined with a single layer of flattened endothelial or mesothelial cells. They are situated in the anterior mediastinum in contact with the anterior thoracic wall, the parietal pericardium and sometimes the diaphragm and one of the lungs.

Cases of cysts of this type have been reported by Dufour and Mourret,[8] Pickhardt[9] and Lambert.[10] The cyst in the case reported by Dufour and Mourret was an incidental observation at autopsy in an 86 year old woman dying of cerebral infarction. Pickhardt reported the case of a 53 year old woman who complained of persistent thoracic pain. At operation a cyst of the anterior mediastinum was encountered and excised, with complete relief of the patient's symptom. Lambert reported 2 cases of cyst in the anterior mediastinum. In each the cyst was asymptomatic and was discovered on roentgen examination in the course of routine studies. In each instance it was removed and the patient had an uneventful postoperative course.

BRONCHIAL CYSTS

Bronchial cysts have also been referred to as bronchogenic, ciliated columnar epithelial and reduplication cysts of the respiratory tract. The theoretic origins include a pinching off of a diverticulum of the foregut in the region of the tracheal bud, a secondary budding of the tracheal bud itself and an abnormal division of the

8. Dufour, H., and Mourret: Bull. et mém. Soc. méd. d. hôp. de Paris **53**:1482, 1929; cited by Lambert.[10]

9. Pickhardt, O. C.: Ann. Surg. **99**:814, 1934.

10. Lambert, A. V. S.: J. Thoracic Surg. **10**:1, 1940.

tracheobronchial tree at a later stage of development.

Origin.—A developmental abnormality seems probable because of the way in which the trachea and the bronchi are formed from the primitive foregut. In the 2.5 mm. embryo the trachea and lung bud are seen as a pear-shaped mass attached to the ventral border of the esophagus. In the 4 mm. embryo the bud begins to bifurcate and the respiratory organs are represented by the laryngeal groove, the tubular trachea and two lung buds or primary bronchi. At this time the cavity in the primitive respiratory organs is still continuous with that of the trachea. The trachea becomes separated from the esophagus by the downward growth of the lung buds and the upward extension of the notch between the lungs and the esophagus. The fusion of the lateral walls to form the tracheoesophageal septum begins from below. It would seem that at this time a pinching off of a small bud or diverticulum of the foregut might occur. This could subsequently be carried caudally by the downward growth of the lungs to the mediastinum. Such a diverticulum would contain entoderm and mesoderm destined to become a part of the trachea, the bronchi, the esophagus, the stomach or the intestine. This theory offers an explanation for bronchial, esophageal, gastric and enteric cysts of the mediastinum.

Structure and Location.—Bronchial cysts may contain any or all of the tissues that are normally present in the trachea and the bronchi. Typically they are lined with ciliated pseudostratified columnar epithelium (fig. 2). In some areas the cilia and even the epithelium may be absent. Their walls consist principally of fibrous connective tissue and may contain mucous glands, hyaline cartilage (fig. 3), smooth muscle, which is sometimes arranged in layers, nerve trunks and elastic fibers. Some of the cysts considered to be bronchogenic are in part lined with stratified squamous epithelium. They vary in size and usually contain clear viscid fluid or gelatinous material. Although they may occur at any site along the tracheobronchial tree, they are most common in the posterior part of the superior mediastinum, particularly in the region of the tracheal bifurcation. The cysts had unusual locations in one of the cases reported by von Westenryk [11] and in that described by Stoeckenius.[12] In both instances the ciliated epithelial cysts were small and unexpected discoveries at autopsy. Von Westenryk's case was that of a 38 year old man who died with right hemiplegia. A small cyst was present in the submucosa of the lower part of the esophagus. Stoeckenius described a similar small cyst within the heart at the upper end of the posterior papillary muscle of the left ventricle. This was an incidental observation in a 45 year old man who died of pneumonia after gastrectomy for carcinoma.

The lumens of the cysts typically do not communicate with the trachea or the bronchi. A communication with the trachea was observed in the 5 cases reported by Chiari.[13] In these instances the anomalies were tracheal diverticula and not true cysts.

Number.—Thirty-four cases [14] of mediastinal cyst of bronchial type had been reported in the literature up to January 1944. To these is added case 2 of this paper, making a total of 35.

Sex and Age of Patients.—Of the 35 patients, 19 were males, and 14 were females; in 2 instances the sex was not indicated.

The distribution of the cases with regard to the age at the time when the diagnosis was confirmed by operation or autopsy is shown in table 3.

Other probable cases have been reported by Harrington,[15] Walzel,[16] Ellis [17] and Stoeckel.[18]

11. von Westenryk: Prag. med. Wchnschr. **25**:373, 1900.

12. Stoeckenius, W.: Zentralbl. f. Herz- u. Gefasskrankh. **11**:73 and 89, 1919; cited by Ehlers.[14o]

13. Chiari, H.: Beitr. z. path. Anat. u. z. allg. Path. **5**:329, 1888.

14. (*a*) Stilling, H.: Virchows Arch. f. path. Anat. **114**:557, 1888. (*b*) Joel, J.: ibid. **122**:381, 1890. (*c*) Zahn, F. W.: ibid. **143**:170, 1896. (*d*) von Rau, F.: ibid. **153**:26, 1898. (*e*) von Wyss, H.: ibid. **51**:143, 1870. (*f*) Eppinger: Pathologische Anatomie des Larynx und der Trachea, Berlin, A. Hirschwald, 1880, p. 256; cited by von Westenryk.[11] (*g*) Herrmann: Prag. med. Wchnschr. **15**:146, 1890; cited by von Westenryk.[11] (*h*) Fletcher, H. M.: Tr. Path. Soc. London **48**:249, 1896-1897. (*i*) von Springer, C.: Prag. med. Wchnschr. **23**:393, 1898; (*j*) Wechsberg, F.: Centralbl. f. allg. Path. u. path. Anat. **11**:593, 1900. (*k*) Bert, P., and Fischer, B.: Frankfurt. Ztschr. f. Path. **6**:26, 1911. (*l*) Blackader, A. D., and Evans, D. J.: Am. J. Dis. Child. **51**:1126, 1936. (*m*) Gold, E.: Beitr. z. path. Anat. u. z. allg. Path. **68**:278, 1921. (*n*) Nossen, H.: Deutsche med. Wchnschr. **51**: 1151, 1925. (*o*) Ehlers, H. W. E.: Deutsche Ztschr. f. Chir. **215**:189, 1928. (*p*) Mixter, C. G., and Clifford, S. H.: Ann. Surg. **90**:714, 1929. (*q*) Alford, J. E.: Arch. Path. **23**:296, 1937. (*r*) Rizzi, I.: Arch. ital. di anat. e istol. pat. **8**:689, 1938; cited by Carlson.[14r] (*s*) Johnston, L. M.: Am. J. Dis. Child. **56**:313, 1938. (*t*) Congenital Cyst of Mediastinum, Cabot Case 26291, New England J. Med. **223**:105, 1940. (*u*) Wyllie, W. G., and Pilcher, R. S.: Arch. Dis. Childhood **18**:34, 1943. (*v*) Carlson, H. A.: J. Thoracic Surg. **12**:376, 1943. (*w*) Adams, W. E., and Thornton, T. F.: ibid. **12**:503, 1943. (*x*) Heuer and Andrus.[4c'] (*y*) Stoeckenius.[12] (*z*) von Westenryk.[11]

15. Harrington, S. W.: Arch. Surg. **19**:1679, 1929.

16. Walzel, P.: Beitr. z. klin. Chir. **158**:654, 1933.

17. Ellis, W. B.: Proc. Roy. Soc. Med. **28**:666, 1935.

18. Stoeckel, K. H.: Zentralbl. f. Gynäk. **59**:2178, 1935.

In these instances the structure was suggestive but not absolutely characteristic of a bronchial cyst.

Symptoms and Signs.—Because of their frequent occurrence in the region of the trachea and the main stem bronchi, bronchial cysts of small size may produce signs of obstruction in the air passages early in life. Not infrequently, however, as in 16 of the 35 cases, the cysts remain quiescent, produce no symptoms and are discovered at autopsy. The cysts were incidentally observed at autopsy in the cases of the 60 year old woman and the 66 year old man reported by Bert and Fischer [14k] and by Rizzi,[14r] respectively, as well as in 14 other instances. Most commonly, however, the symptoms appear during the first

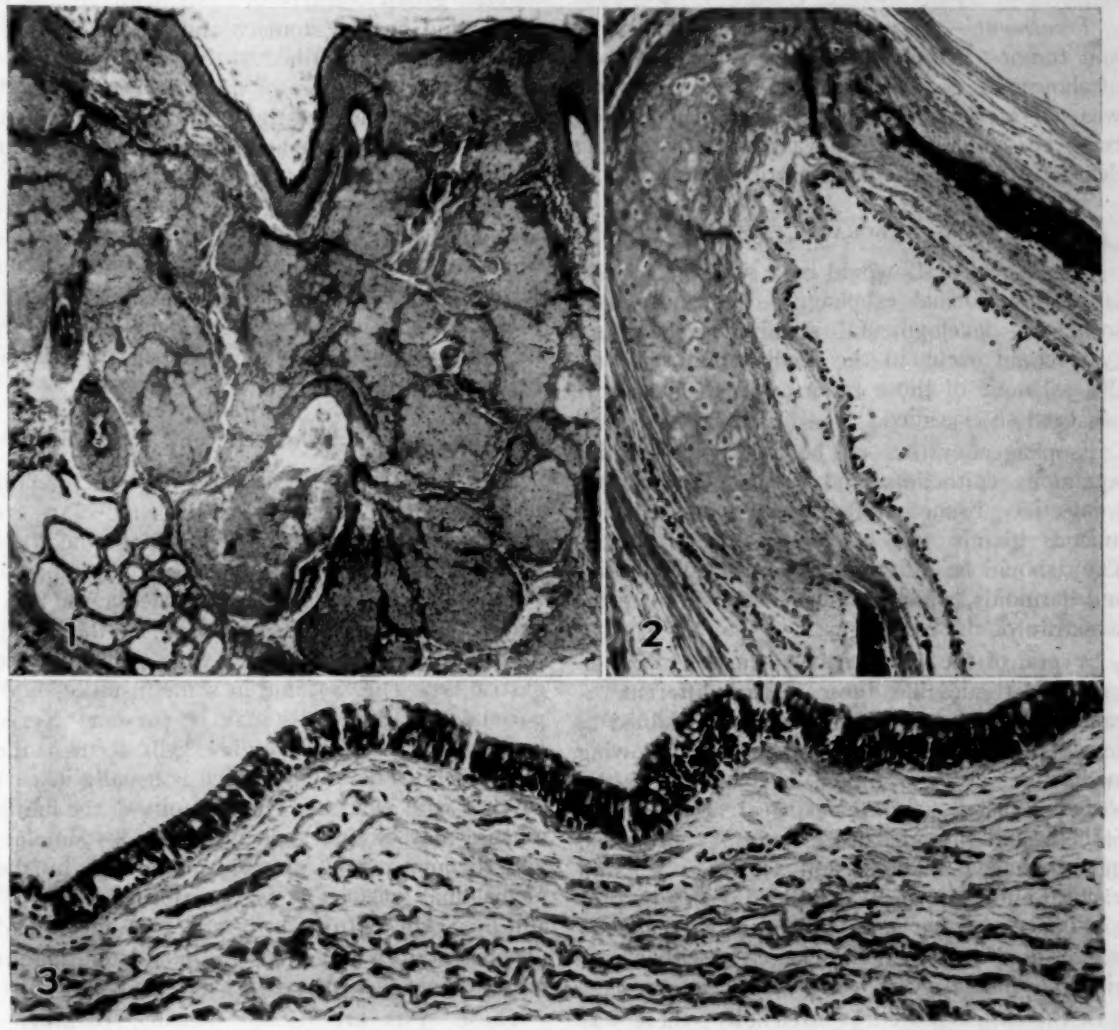


Fig. 1 (case 1).—Stratified squamous epithelium lines the cyst. Hair follicles, apocrine glands and many sebaceous glands are in adjacent connective tissue. Hematoxylin and eosin; × 385.

Fig. 2 (case 2).—Typical bronchial or ciliated pseudostratified columnar epithelium lines the cyst. Hematoxylin and eosin; × 158.

Fig. 3 (case 2).—Cartilage and bone are present in the cyst wall. The inner part of the lining ciliated pseudostratified columnar epithelium is desquamated. Hematoxylin and eosin; × 117.

TABLE 3.— *Age Incidence*

Age Group	Number
Stillborn	1
Less than 1 year	9
1 to 9 years	3
10 to 19 years	1
20 to 29 years	3
30 to 39 years	7
40 to 49 years	4
50 to 59 years	2
60 to 69 years	2
Age not stated	3

few months of life. It is even possible that the infant with such a cyst may be stillborn because of it. Cyanosis, dysphagia (difficulty in feeding) and cough are also frequent.

Examination may reveal signs of pneumonia or atelectasis of the lung. Dulness to percussion, cardiac displacement, bulging of the thorax and scoliosis may be noted. The diagnosis during

life may be difficult or impossible to reach because of the small size and the posterior position of the cyst. Such cysts are difficult to visualize by roentgen rays, especially when, as is commonly true, they are surrounded by atelectatic or consolidated pulmonary tissue. Bronchography and especially bronchoscopy should be helpful in diagnosis by demonstrating the narrowing of the trachea or of the bronchi due to extrinsic pressure.

Treatment.—As with other mediastinal cysts and tumors, complete excision is the treatment of choice. In 6 of the reported cases the patient was operated on. The cyst in 4 was completely removed, and the patient was cured; in 2 cases death resulted from the operation.

ESOPHAGEAL CYSTS

Esophageal cysts would have structures simulating the normal esophagus. It seems likely from the developmental standpoint that such cysts could occur in the mediastinum. Nevertheless, none of those reported in the literature has been so classified.

Esophageal cysts would be lined with stratified squamous epithelium and would have fibrous connective tissue walls, possibly containing mucous glands and layers of smooth muscle. They should be differentiated from epidermoids and dermoids, which do not closely simulate the structure of the esophagus.

Several of the cases reported in the literature have had tissues like those of two different organs. Such mixed cysts are easily explained as developmental abnormalities. The following mediastinal cysts of mixed type have been reported: gastric and esophageal (Carlson [4k]; Stähelin and Burckhardt [19]; Smith [20]), bronchial and esophageal (Adams and Thornton [14k]) and tracheal and esophageal (Guillery [21]). The predominance of either gastric or bronchial elements has resulted in their being classed as either gastric or bronchial cysts.

In the case reported by Melchoir [22] there was a mediastinal cyst with a microscopic structure much like that of the normal esophagus. The lining epithelium was almost entirely of the stratified squamous type and the fibrous connective tissue wall contained mucous glands. A single focus of columnar epithelium was noted, but the presence of cilia could not be established with certainty. The author considered this to be a bronchial cyst, the original bronchial epithelium having undergone metaplasia to the stratified squamous type. Such an occurrence certainly cannot be denied but it also seems possible that the pinching off of a portion of foregut made up of tissue destined to be esophageal could have occurred.

GASTROENTERIC CYSTS

This group consists of cysts with structures simulating normal stomach and intestine. Their origin has been ascribed to the pinching off of a bud or a diverticulum of the embryonic foregut, to an intrathoracic vestige of the omphalomesenteric (vitelline) duct and to proliferation of an entodermal germ cell of the esophagus.

Occasionally the gastric and enteric cysts are structurally so similar that careful examination of several sections from different portions of the wall is necessary for proper classification. As indicated in an earlier section, some of the cysts in this group also contain esophageal, tracheal or bronchial elements.

GASTRIC CYSTS

Structure and Location.—The microscopic features of the gastric or gastrogenic cysts are like those of normal stomach. All of the coats of normal stomach may be present: the mucosa, the muscularis mucosae, the submucosa, the muscularis and the serosa (fig. 4). In the lining mucosa there are deep branching glands of the gastric type (fig. 5), and in some instances both parietal and chief cells may be present. Nerve trunks and groups of ganglion cells occur in the muscularis. The outer surface is usually in part covered with pleura. The continuity of the lining mucosa may be interrupted, and lesions simulating chronic peptic ulcers were noted in the mediastinal gastric cysts described by Böss [23] and by Seydl.[24] In both instances the ulcers had penetrated into the adjacent lung, resulting in intrapulmonary hemorrhage and hemoptysis.

These cysts are usually situated paravertebrally in the posterior mediastinum, behind the trachea and the esophagus. They vary considerably in size, are usually unilocular and contain from a few to several hundred cubic centimeters of clear amber, turbid or sanguineous viscid fluid. This fluid may be neutral or strongly acid in reaction. The cysts are frequently firmly adherent to adjacent structures, especially the esophagus and the lung. Attachment to or extension into the diaphragm and erosion of vertebrae

19. Stähelin and Burckhardt: Arch. f. Verdauungskr. **15**:584, 1909; cited by Seydl.[24]

20. Smith, R. E.: Guy's Hosp. Rep. **80**:466, 1930.

21. Guillery, H.: Zentralbl. f. allg. Path. u. path. Anat. **69**:49, 1937.

22. Melchoir, E.: Zentralbl. f. Chir. **56**:2626, 1929.

23. Böss, C.: Virchows Arch. f. path. Anat. **300**:166, 1937.

24. Seydl, G. N.: Frankfurt. Ztschr. f. Path. **52**:346, 1938.

and ribs were noted in the cases reported by Mixter and Clifford.[14p]

Number.—Twelve cases [25] of mediastinal gastric cyst had been recorded in the literature up to Jan. 1, 1944. To these are added the case recently reported by Olken [26] and case 3 of this paper, making a total of 14 such cases.

Sex and Age of Patients.—Of the 14 patients, 8 were males, and 5 were females; in 1 case the sex was not stated.

All the patients were infants or children, as indicated in the following tabulation:

	Number
Less than 1 year	7
1 to 2 years	4
2 to 10 years	3

Symptoms and Signs.—The symptoms, like those of other similarly located cysts, are largely due to compression of the trachea or the bronchi and include dyspnea, cyanosis, dysphagia (difficulty in feeding) and occasionally, even in infants,[24] hemoptysis.

The physical findings may be primarily those of pulmonary atelectasis or pneumonitis. Cardiac displacement, scoliosis and bulging of the chest also occur. Although the contents may be neutral in reaction, the aspiration of strongly acid fluid should be helpful in diagnosis.

Treatment.—In 6 of the 14 cases complete surgical excision was attempted. In 3 complete excision cured the patient; in the other 3 death occurred during or soon after the operation.

ENTERIC CYSTS

These have also been called enterogenous cysts, *Enterokystome, Darmkystome* and *Darmzysten* (intestinal cysts). They are similar to the gastric cysts except that their microscopic structure simulates that of the normal intestine.

Number.—Three cases with such mediastinal cysts [27] had been recorded in the literature up to Jan. 1, 1944. Two other probable cases of enteric cyst have been reported.[28] The absence of mucosa in the cyst in each case prevents exact classification. Both cases should, therefore, be designated as cases of gastroenteric cyst of the mediastinum.

Sex and Age of Patients.—Four of the 5 patients were males, and 1 was a female. Three were stillborn, 1 was 15 months old and 1 was 4½ months of age.

Intra-abdominal cysts of similar type were present in 3 of the 5 cases of enteric or probable enteric cyst. The concomitant occurrence of such cysts in the abdomen and the thorax suggests the possibility of their originating from the omphalomesenteric duct.

CYSTIC LYMPHANGIOMA

Cystic lymphangioma (cystic hygroma) is probably of congenital origin and is among the less common of the mediastinal cystic tumors. It is characterized by multiple variable-sized cystic spaces, lined with a single layer of endothelium and containing gelatinous material or fluid that is clear and colorless or brown. The fibrous connective tissue between the spaces not infrequently contains scattered smooth muscle fibers, foci of lymphocytes, fat cells and cholesterol crystals. The lesion is not encapsulated and has ill defined borders, usually intimately associated with the great vessels and surrounding structures, thus making complete excision almost impossible. Attempts at removal may result in profuse bleeding.

The mode of origin is not certain. It is suggested that a portion of the anlage for the formation of blood vessels is drawn down from the gill clefts by the descent of the pericardium. An origin from the thymus has also been considered. In some cases the mediastinal cystic lymphangioma may be the result of direct intrathoracic extension of a similar tumor of the neck.

Cases of mediastinal lymphangioma have been reported by Michaelis [29] and by Skinner and Hobbs.[30] The symptoms are not pathognomonic of this type of cyst but the onset of symptoms is probably earlier than in others.

REPORT OF CASES

CASE 1.—R. P., a white woman 22 years of age, was admitted to the hospital on June 16, 1944. She had been in good health until several months prior to hospitalization, when she contracted lobar pneumonia; a roentgenogram of the chest revealed a large tumor of the mediastinum. Several roentgen treatments were given over the site of the tumor. It apparently decreased in size for a short time and then began to

25. Entz, B., and Orosz, D.: Frankfurt. Ztschr. f. Path. **40**:229, 1930. Fischer, W.: Virchows Arch. f. path. Anat. **275**:711, 1930. Poncher, H. G., and Milles, G.: Am. J. Dis. Child. **45**:1064, 1933. Nicholls, M. F.: Brit. J. Surg. **29**:137, 1940. Mixter and Clifford.[14p] Carlson.[4k'] Wyllie and Pilcher.[14u] Guillery.[21] Stähelin and Burckhardt.[19] Smith.[20] Böss.[23] Seydl.[24]

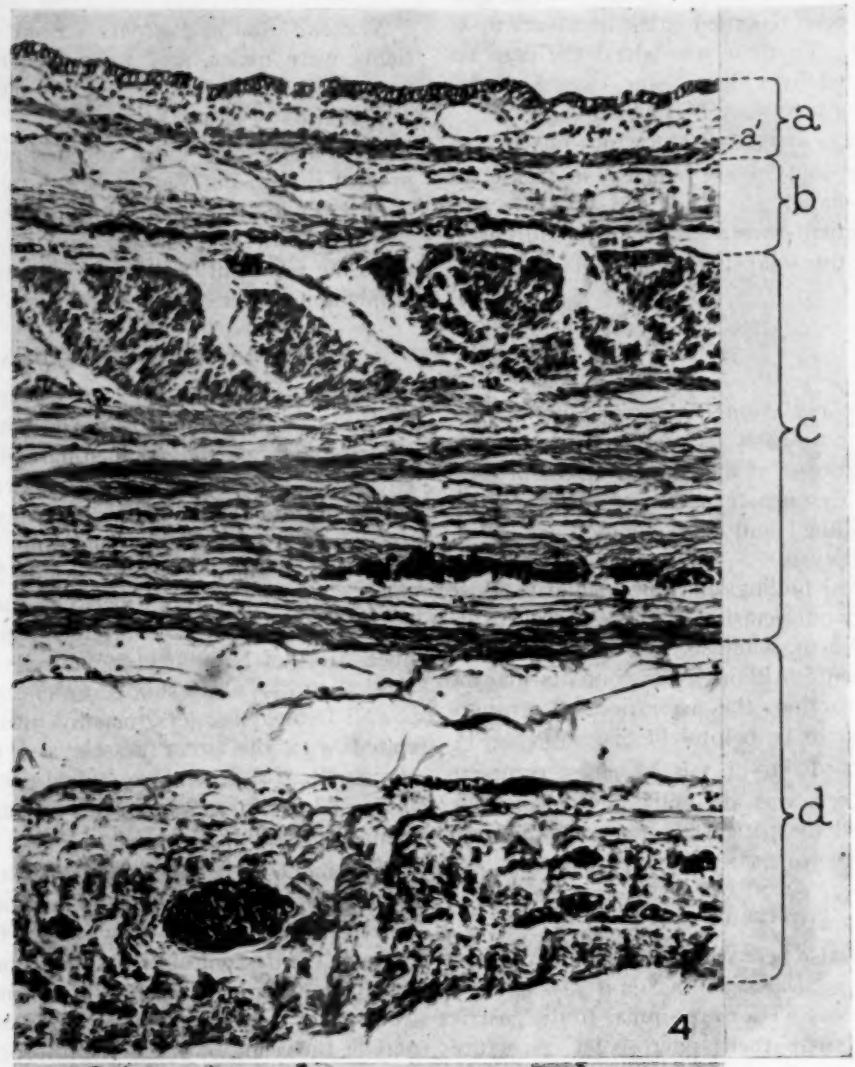
26. Olken, H. G.: Am. J. Path. **20**:997, 1944.

27. Roth, M.: Virchows Arch. f. path. Anat. **86**:371, 1881. Brass, K.: Frankfurt. Ztschr. f. Path. **50**:26, 1936. Schminke, A.: Virchows Arch. f. path. Anat. **227**:12, 1920.

28. Hennig, C.: Centralbl. f. Gynäk. **4**:398, 1880. Black, R. A., and Benjamin, E. L.: Am. J. Dis. Child. **51**:1126, 1936.

29. Michaelis, O.: Deutsche Ztschr. f. Chir. **242**:250, 1934.

30. Skinner, G. F., and Hobbs, M. E.: J. Thoracic Surg. **6**:98, 1936.



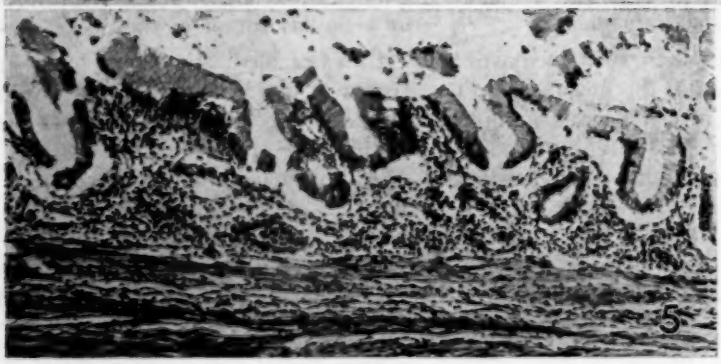


Fig. 4 (case 3).—Four coats like those of the intestine are present in tne cyst wall: (*a*) mucosa; (*a'*) muscularis mucosae; (*b*) submucosa; (*c*) muscularis (two layers); (*d*) serosa. Hematoxylin and eosin; × 165.

Fig. 5 (case 3).—Glands like those of gastric mucosa are in the inner part of the cyst. Hematoxylin and eosin; × 102.

enlarge relatively rapidly. During the nine months subsequent to her recovery from pneumonia the patient had a persistent cough, productive of thick yellow sputum. The cough was worse when she was in the recumbent position. Blood was coughed up on two occasions. For two weeks before hospitalization she had had constant substernal pain.

On admission she was normally developed and well nourished. The body temperature was 37.8 C. (100 F.), the pulse rate 100, the respiratory rate 25 and the blood pressure 130 systolic and 90 diastolic. The thorax was symmetric and normally resonant, with diminished breath sounds to the right of the midline posteriorly at the level of the fourth to the sixth thoracic vertebrae. The urine was normal; the blood count showed 8,600 white cells. The hemoglobin was 75 per cent (Sahli). Roentgenograms showed a lobulated, sharply defined tumor (12 by 5 by 6 cm.) along the right side of the heart. In its upper part were several molar teeth and an irregularly shaped mass of bone. A diagnosis of teratoma was made.

On her third day in the hospital the patient was operated on. The anterior portion of the right fourth rib was resected, and a large cystic tumor was seen in the anterior mediastinum, about 10 cm. in diameter. The tumor was firmly adherent to the upper lobe of the right lung, from which it was dissected with considerable difficulty. The outer portion of the cyst wall could not be completely excised, but all of its inner portion and contents were removed. The lung was cut into and sutured at several sites. A drainage tube was inserted. For nine days after the operation the patient's temperature fluctuated between 38.2 and 41 C. (100.7 and 105.8 F.). The white blood cell count varied between 14,600 and 13,200. Over a period of eleven days following the operation 1,275,000 units of penicillin were administered. After this the patient gradually improved, and her temperature returned to normal. She was discharged on the seventeenth postoperative day.

The surgical specimen consisted of four irregularly shaped pieces of tissue, weighing 205 Gm., evidently portions of the wall of a cyst. There was a large piece of bone with two attached molar teeth, considerable grumous material in which there were many brown hairs, some gray fibrous tissue and yellow fat. Microscopic examination showed the cyst to have a stratified squamous epithelial lining. The wall consisted of fibroadipose tissue of mature type, and in its inner portion there were many hair follicles and sebaceous glands (fig. 1). Attached to one part was a small piece of lung, the seat of marked acute and chronic interstitial pneumonitis.

Diagnosis: Dermoid cyst of the anterior mediastinum.

Case 2.—J. M., a Negro man aged 45 years, was hospitalized because of painless jaundice and marked loss of weight. An exploratory laparotomy revealed tumor in the peripancreatic tissue, the omentum and the liver. The tissue was unusually friable, and an artery was inadvertently torn. Shortly after the profound hemorrhage which resulted, the patient went into shock and died.

Autopsy (Dr. O. Eitzen) revealed well differentiated scirrhous adenocarcinoma of the tail of the pancreas with metastases in the liver, the peritoneum and the abdominal lymph nodes and extension into and partial obstruction of the main hepatic bile duct. There were also laceration of the gastroduodenal artery, generalized icterus and cholemic nephrosis.

An incidental finding was a cyst in the superior mediastinum, measuring 6 cm. in diameter. It was attached to the superior surface of the heart, posterior and to the left of the ascending aorta. It was firmly adherent to the outer surfaces of the atriums, the parietal pericardium, the trachea and the upper portions of the lungs. Its wall consisted of firm pale gray tissue and varied from 1 to 2 mm. in thickness. The inner surface was in large part glistening and pale gray, but at some sites it was rough and yellowish brown. The contents consisted of viscid turbid dark brown fluid which solidified on fixation in 4 per cent solution of formaldehyde.

Microscopically, the cyst was lined with ciliated pseudostratified columnar epithelium. In the roughened areas the epithelium was absent and there was hemosiderin in the inner part of the wall, indicative of remote hemorrhage. The wall consisted of fibrous connective tissue in which there were hyaline cartilage, spicules of bone and groups of smooth muscle fibers (fig. 3).

Diagnosis: Bronchial cyst of the superior mediastinum.

Case 3.—The patient was a premature white boy with a body weight of 1,500 Gm. and a length of 38 cm. In the lower thoracic and lumbar regions there was a large defect of the skin, subcutaneous tissue and dorsal portions of the vertebrae. Death occurred twenty-seven hours after birth.

Autopsy (Dr. B. Chomet) showed a cyst in the posterior mediastinum, measuring 3 by 2.5 by 2 cm. It contained light brown fluid and had a glistening inner surface and pale gray wall, 1 mm. in thickness. The cyst was immediately adjacent to but not connected with the esophagus. Microscopic examination revealed distinct coats like those of the intestine (fig. 4). The outer dense fibrous connective tissue layer was like intestinal serosa. Adjacent to this was a muscular coat with an outer circular and an inner longitudinal layer. Submucosal and mucosal coats were also evident, with a thin muscularis mucosae. The lining epithelium was largely of simple columnar type, but in some regions (fig. 5) there were high columnar epithelial cells and glands of a mucus-secreting type.

Diagnosis: Gastric cyst of the posterior mediastinum; spina bifida, umbilical hernia, Meckel's diverticulum and fetal atelectasis of lungs.

SUMMARY

The great variety of mediastinal cysts and cystic tumors may be classified as congenital (six types) and acquired (three types). A review of the literature reveals a large number in the congenital group. Additional dermoid, bronchial and gastric cysts of the mediastinum are reported in this paper. With these there are recorded in the literature 245 epidermoids, dermoids and teratomas, 35 bronchial cysts, 14 gastric cysts and at the most 5 enteric cysts of the mediastinum. Eleven and four-tenths per cent of the epidermoids, dermoids and teratomas were cancerous. All other types of congenital mediastinal cysts were noncancerous. Such cysts and tumors may give rise to symptoms late in life but the majority have reached sufficient size to cause symptoms during the first three decades. The most common symptoms were cough, pain in the chest, dyspnea and hemoptysis. The treatment of choice is complete surgical excision—if possible, prior to the development of pressure symptoms or infection.

SOME ENDOCRINOLOGIC CONSIDERATIONS OF CANINE NEOPLASTIC DISEASES

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The histologic study of a number of canine neoplasms observed at autopsy and in surgical specimens and some preliminary success in the production of neoplasms in dogs by means of carcinogens stimulated a review of the available literature on canine oncology in an attempt to assemble pertinent facts to form a base line for future investigation in this field. Of particular interest in the publications surveyed was much material concerned with neoplasms of endocrine glands and of the male and female genital tract. The main headings under which the various endocrinologic considerations of canine neoplastic diseases will be taken up are as follows: "Neoplasms of Endocrine Glands Associated with Endocrinic Disturbances," "Neoplasms of the Male Genital Tract," "Neoplasms of the Female Genital Tract," "Neoplasms of Other Endocrine Glands," "Infectious or Venereal Sarcoma" and "Mammary Neoplasms."

NEOPLASMS OF ENDOCRINE GLANDS ASSOCIATED WITH ENDOCRINIC DISTURBANCES

A syndrome of feminization in male dogs associated with carcinoma of the testis and mimicked following the administration of estrogens was recently summarized [1] as it occurred in 6 male dogs described in the literature. These animals possessed some or most of the following features: (1) adenocarcinoma of the testis, (2) varying degrees of atrophy of the opposite testis, (3) stratified squamous epithelial metaplasia of the prostatic urethra, ducts and acini with hypertrophy of the prostate, (4) hyperplasia of the ducts and acini of the breasts with mammary enlargement, (5) swelling of the penile sheath, (6) loss of hair, (7) attraction of other male dogs, like a female dog in estrus, and (8) depression of libido. Two other cases probably related to the 6 already reviewed include case 2 of Baldoni [2a] and that of a cryptorchid dog described by Krause.[3] Baldoni's case 2 was that of a male hound-pointer cross, about 10 years old, which in the course of nine months displayed gradual increase in the size of the right testis and later enlargement of all breasts. The right scrotal testis was as large as a fist, the left testis was small and there were tumors in the fourth and fifth breasts. Histologic examination of the surgically removed tumors indicated that the testicular neoplasm was a carcinoma (seminiferous epithelioma or seminoma of Chevassu) and that all four mammary neoplasms were adenocarcinomas. The histologic characteristics of the testicular tumor were the same as those of the carcinomas in ectopic testes described by the author in 1913 and 1923.[2b] Neither of these articles was obtainable for first hand perusal. Baldoni's case 1 was that of a male pointer, about 7 years old, which for seven months had a tumor of the right hindmost breast, which gradually enlarged to the size of a fist. The animal had always showed strong sexual desire, but with the appearance of the mammary tumor it had become more retiring and remained indifferent to the presence of bitches in heat. His stamina in the hunt and his general conduct were unaltered. His testes were firm and slightly tender. The excised mammary tumor was histologically a fibroadenoma. Worthy of mention in this case, according to Baldoni, was the coincidence of disappearance of strong sexual desire, an indication of testicular atrophy, with the start of the development of the mammary fibroadenoma. Baldoni stated that it had been demonstrated in man that the breasts may be stimulated after traumatic testicular atrophy (Pende; Falta), iodine intoxication (Bergess) or other types of intoxication, like hepatic cirrhosis (Silvestrini; Corda; Zanalda; Monai; Tattoni) and that sometimes after extirpation of the testes in adults the mammary glands increase in size (Tobler) and even become functional. Baldoni mentioned a Swiss bull which soon after castration showed a remarkable enlargement of the breasts and production of milk. Krause [3] described a male hound

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1. Mulligan, R. M.: Am. J. Path. **20**:865, 1944.

2. Baldoni, A.: Mem. r. Accad. d. sc. d. Ist. di Bologna (*a*) **5**:33, 1927-1928; (*b*) **10**:183, 1912-1913; **10**:37, 1922-1923.

3. Krause, C.: Frankfurt. Ztschr. f. Path. **41**:405, 1931.

8 years old, which six years before had been submitted to orchidectomy of the left side because of excessive sexual urge. After this operation he was never known to have had coitus again. Autopsy revealed an enlarged right testis in the right iliac region. The left testis (presumably formerly scrotal) was operatively absent. Also observed were a greatly enlarged, cystic prostate, a hypertrophied urinary bladder and a thickened urethra. The anatomic diagnoses after histologic study were: alveolar carcinoma of the right testis, carcinoma of the prostate, hypertrophy of the urinary bladder, chronic pyelonephritis and atrophy of the right epididymis and ductus deferens. The anatomic description and photomicrographs of the prostate when carefully perused indicate that the true condition of this gland was stratified squamous epithelial metaplasia of the prostatic urethra, ducts and acini and not carcinoma. The hypertrophy of the urinary bladder and the chronic pyelonephritis were probably the results of prostatic enlargement and urethral obstruction. The diagnosis of carcinoma of the right intra-abdominal testis in this case is unquestionably valid from the evidence presented. Zuckerman and McKeown[4] reported observations on a group of 15 dogs with testicular adenocarcinoma. Five of these dogs showed concomitant squamous metaplasia of the gland system of the prostate and a sixth dog had an associated prostatic carcinoma. The authors presumed that estrogenic hormone had been elaborated by the adenocarcinoma in the 5 dogs with squamous metaplasia of the prostate, a finding indicating estrogenic stimulation. In summary, it may be said that male dogs suffering from testicular carcinoma may show certain clinical abnormalities and tissue changes, which have been detailed; the basis for these is some sort of estrogenic stimulation through a substance elaborated directly by the testicular tumor or of some substance secreted by the tumor and changed by a metabolic process into a compound with a feminizing action.

Although not strictly a neoplasm but rather a form of hyperplasia, benign prostatic hypertrophy (which commonly occurs in old male dogs as demonstrated in 22 of 37 male dogs afflicted with this lesion in the series of Goodpasture[5]) may be influenced to the point of regression by castration. Hobday[6] described 3 male dogs in which prostatic hypertrophy was relieved by castration. The first was a toy terrier, about 4 years old, with strangury, oliguria and prostatic enlargement so severe that a catheter could not be passed. Within three weeks after castration, performed after the animal had been anesthetized with chloroform, urination was free, a catheter was passed easily, the prostate was smaller, and other symptoms were absent. The second was a collie, 9 years old, with straddling gait of the hindlegs, difficulty in walking or rising, objective pain in the loins and flanks, and difficulty in starting his stream, with subsequent passage of a large amount of urine. Ten days after castration, urine was passed easily, and the formerly enlarged prostate was much smaller. The third was a Dandie Dinmount, about 11 years old, with dysuria, straddling gait and an enormous prostate, painful to pressure. Three days after castration the urinary flow was unimpeded; no relapse was observed two years later. These cases indicate that castration either breaks a chain controlled by the pituitary gland or eliminates the interstitial cells of the testes as a primary source of androgenic stimulus. The reason that castration works so well in alleviating prostatic hypertrophy in the dog as compared with orchidectomy in men afflicted with this malady may be contained in a paper by Moore,[7] who stated that 80 per cent of male dogs over 8 to 10 years of age show enlargement of the prostate. His opinion was that the etiologic factors may be the same in both man and the dog but that the essential lesion in prostatic enlargement in the dog is diffuse hyperplasia while that in man is nodular hyperplasia. In the experience of Zuckerman and McKeown,[4] diffuse hyperplasia was the rule in enlargement of the canine prostate. In the human prostate affected by benign enlargement one can demonstrate hyperplastic glandular nodules, atrophic cystic epithelial structures, mostly peripherally placed, often distended by inspissated material and involved in inflammatory changes, and proliferated fibromuscular stroma, sometimes arranged in the form of actual fibroleiomyoma and often marked by foci of chronic inflammatory cells. Because of these exaggerated anatomic abnormalities inherent in nodular hyperplasia (if not for some unknown hormonal factors) of the human prostate, it seems logical that after castration they would be less likely to regress than the rather symmetrically proliferated prostatic structures observed in the diffuse hyperplasia characterizing benign enlargement of the canine prostate.

4. Zuckerman, S., and McKeown, T.: J. Path. & Bact. **46**:1, 1938.

5. Goodpasture, E. W.: J. M. Research **38**:127, 1918.

6. Hobday, F. T. G.: Surgical Diseases of the Dog and Cat, edited by J. McCunn, Baltimore, Williams & Wilkins Company, 1939, pp. 273-274.

7. Moore, R. A.: Surgery **16**:152, 1944.

The papers cited [8] described endocrine changes in association with tumors of the canine male sex glands. The first discovered instance of endocrine changes in association with tumors of canine female sex glands was described by Wolfe, Cleveland and Campbell.[9] With the genitalia as a guide to the stage in the estrual cycle at which the gland was examined, these authors quantitatively studied the cellular composition of the anterior lobe of the pituitary gland in 79 female dogs. One animal which showed external bleeding ten days before autopsy disclosed follicular cysts of the ovaries and cystic glandular hyperplasia of the endometrium. The anterior lobe of the pituitary gland of this dog contained an increased number of acidophils. DeVita,[10] basing his preliminary report on the tissues and histories of 24 cases, in 5 of which histologic sections were made, described the clinical features, the ovarian and uterine pathologic changes and illustrative cases of the condition known as hyperplastic endometritis or so-called pyometra of the bitch. He also quoted Lesbouyries and Berthelon, but listed no specific reference to their work on 4 dogs with the disease which were treated with gonadotropic hormone and oophorectomy. DeVita stated that hyperplastic endometritis may be seen following completion of an estrual cycle at any age but is most often observed in dogs over 6 years old. Virgins as well as parous females are afflicted with the disease, the most common cause of sterility in the bitch. The course may be chronic and intermittent or may end suddenly and unexpectedly in death. The clinical features include: malaise; fever; prolongation or shortening of the estrual cycle with increase or decrease in the physical evidences of estrus; vulvar hypertrophy with swelling, hardening and secondary traumatic changes; gray-yellow to red-brown, usually putrid vaginal discharge; abdominal distention due to the enlargement of the uterus, simulating pregnancy; mammary tumors and sometimes inguinal hernia; attacks of lumbago; straddling gait of the hindlegs; dry, inelastic skin; sparse, dry, brittle hair, and varying degrees of alopecia with pigmentation especially on the dorsum, the sacrum, the flanks, the abdomen, the perineum and the pudendum. In the ovaries the following abnormalities were observed: cystic follicles, either solitary or multiple; tumors ranging from fibroma to epithelioma in 5 of the 24 cases; corpora lutea fairly fresh in early cases and variously regressed in chronic cases; and all stages of follicle growth and regression. In the early phases, the uterus was enlarged, hyperemic, softened, distended and plicated, the cervix was small, and the ovaries contained fresh corpora lutea. In the later phases varying fibrosis, endometrial and serosal cysts, diverticula, solitary or multiple tumors and thickened cystic endometrium were present. Histologically, the endometrium displayed extreme glandular hyperplasia with edema and hyperemia, followed by retrogressive changes including necrosis, cystic change, abscess formation and fibrosis. Projections of hyperplastic endometrium extended into the myometrium which was involved by hyperemia, edema and fibrosis, which also affected the cervix.

DeVita divided the cases of hyperplastic endometritis into two varieties. The first included those cases in which a metestrual type of pyometra or uterine abscess occurred and those in which this condition (because of the presence or reestablishment of cervical drainage) went into retrogression and some regression, to become activated again by completion of another cycle. Illustrative of the first point was a cocker spaniel bitch, 13 years old, which over a period of three years had several attacks of pyometra following completion of an estrual cycle or occurring at a time when an estrual cycle might have been completed. Bilateral oophorectomy and partial right uterine cornuectomy showed a cystic condition of both ovaries, which contained corpora lutea, and cystic glandular hyperplasia of the endometrium. At autopsy, three weeks later, the hyperplastic endometrial changes in the remnant of the right uterine horn had greatly regressed. Emphasizing the second point was the case of an old poodle with reactivated endometritis, many breeding failures, frigidity and shortening of the interestrual interval. The uterus showed fibrosis, extensive cystic change, edema, hyperemia and focal abscesses. The vagina was fibrotic and lined by papillary stratified squamous epithelium. The second variety of cases included those in which the hyperplastic endometritis was associated with ovarian tumor. These were typified in the case of a cocker spaniel bitch, 6 years old, which was mated in estrus to one dog and fourteen days later to another but did not whelp to either. She again accepted a male fifty-nine days after the first breeding. She was in a constant state of nymphomania from this time until one hundred and fourteen days later when panhysterectomy revealed a chronic fibrotic hyperplastic condition of the uterus and ovarian tumors.

In discussing the etiologic factors of hyperplastic endometritis in the dog, DeVita indicated that estrogen-producing ovarian cysts and tumors appear to be a probable source of the inciting

8. Mulligan.[1] Baldoni.[2] Krause.[3] Zuckerman and McKeown.[4] Goodpasture.[5] Hobday.[6] Moore.[7]

9. Wolfe, J. M.; Cleveland, R., and Campbell, M.: Ztschr. f. Zellforsch. u. mikr. Anat. **17**:420, 1933.

10. DeVita, J.: J. Am. Vet. M. A. **95**:50, 1939.

factor when associated with some corpus luteum activity. This would also serve to explain the uterine changes in the dog reported on by Wolfe, Cleveland and Campbell.[9] In view of the abnormal cellular composition of the anterior lobe of the pituitary gland of this animal, future studies on "hyperplastic endometritis" might profitably be concerned with a histologic analysis of all the endocrine glands and secondary sex glands as well as of the parenchymatous viscera.

The only other neoplasm of an endocrine gland causing hormonal effects was reported by Slye and Wells[11] as occurring in a female white pit bull terrier, 12½ years old, which during life exhibited evidences of hypoglycemia, with low values for blood sugar, and the ability of knowing when to ingest carbohydrate for the relief of symptoms. At autopsy this animal had an adenocarcinoma of the pancreas with metastases in the peripancreatic lymph nodes, a benign adenoma of the pancreas, a mixed tumor of the right posterior mamma, 2 subcutaneous lipomas of the abdominal wall and a sebaceous adenoma of the forefoot. Another case of pancreatic cancer in a male dog was described by Bru,[12] but the only symptom listed was dyspnea. The pancreas was thickened and dense and was sprinkled with firm, white-yellow nodules consisting of islet tissue in the form of irregular palely stained sheets of cells with a rich vascular supply. The structure of the metastases in the liver, the bronchial lymph nodes, the right auricle of the heart and the lungs was much like that of the primary tumor.

NEOPLASMS OF THE MALE GENITAL TRACT

Künnemann[13] reviewed the published descriptions of four testicular tumors in dogs and added a report of 8 of his own. Siedamgrotsky[14] reported the case of a 17 year old male dog with medullary sarcoma of the testis and metastases in the retroperitoneal lymph nodes. Pauer[14] found a carcinoma in the retroperitoneal testis of a 9 year old dog. Calvé[14] observed a dog with a primary carcinoma of the testis and a metastasis in the liver. In a dog over 10 years old Duschanek[14] observed a large cell alveolar sarcoma in retroperitoneal testes bound to the lumbar vertebrae. Künnemann described 8 dogs with neoplasms as follows: a pinscher, 9 years old, with a carcinoma of the left inguinal testis; a setter, 11 years old, with a sarcoma of the right scrotal testis; a Dalmatian, about 1 year old, with a sarcoma of the left scrotal testis; a spitz, 10 years old, with a sarcoma of the right inguinal testis; a terrier with a fibrosarcoma of the right scrotal testis; a dachshund, 6 years old, with an interstitial cell tumor of the left scrotal testis; a pinscher, about 8 years old, with an interstitial cell tumor of the right testis, and an old poodle with an interstitial cell tumor of the right testis. Goodpasture[5] reported 4 cases of carcinoma of the canine testis. In each instance the tumor had a multicentric origin from the regressed epithelium of the tubules. In 1 case the tumor metastasized to a pelvic lymph node.

Ball[15] recorded 2 cases of interstitial cell tumor of the testis; in both the seminiferous tubules were pushed aside and showed compression atrophy. Kunze[16] observed 13 dogs, 9 to 15 years old, with solitary or multiple, unilateral or bilateral testicular interstitial cell tumors, which varied from the size of a pepper grain to that of a small hazelnut. This author found that such new growths occurred often in the atrophic testes of old dogs, always remained within the confines of the tunica albuginea, caused no testicular enlargement and were well defined yellow-white nodules in their early development but in later stages showed hemorrhagic necrosis and encapsulation by connective tissue. A rich network of capillaries and little connective tissue were present within the tumors, the lipoid content of the cells of which could be strikingly and characteristically demonstrated with the sudan IV stain. In a series of 15 old dogs, most of them over 10 years old, Bouffannais[17] observed 2 grossly visible testicular tumors, a seminoma and an interstitial cell tumor. Chambers[18] saw a fox terrier, 8 years old, with a sarcoma of the testicle.

Pallaske[19] studied the organs of 107 male dogs at autopsy. Of the 31 with so-called interstitial cell tumors of the testes, 9 were under 10 years of age and the rest were 10 to 16 years old. In 28 of the dogs with these testicular tumors he also found nodular hyperplasia of one or more of the following organs: liver, adrenal glands, spleen, pancreas and thyroid gland. This study emphasized the difficulty in a given case of interstitial cell growth of the testis of an old dog of

11. Slye, M., and Wells, H. G.: Arch. Path. **19**:537, 1935.

12. Bru, P.: Rev. méd.-chir. d. mal. du foie **2**:40, 1927.

13. Künnemann, O.: Arch. f. wissensch. u. prakt. Tierh. (supp.) **36**:229, 1910.

14. Cited by Künnemann.[13]

15. Ball, M.: Bull. Assoc. franç. p. l'étude du cancer **11**:5, 1922.

16. Kunze, A.: Virchows Arch. f. path. Anat. **240**: 144, 1923.

17. Bouffannais: Bull. Assoc. franç. p. l'étude du cancer **18**:808, 1929.

18. Chambers, F.: Vet. Rec. **11**:709, 1931.

19. Pallaske, G.: Virchows Arch. f. path. Anat. **281**:856, 1931.

determining whether the proliferated interstitial cells might constitute merely hyperplasia or true neoplasm and, once the neoplastic character of such a growth was established, whether adenoma or carcinoma might be the correct diagnosis. Among 1,111 dogs examined at autopsy Skoda [20] found 21 that had neoplasms of the testes; 14 of these dogs had abnormally and 7 normally located testes. On the other hand, Lehr [20] studied 969 surgically removed canine tumors, among which were 34 (3.5 per cent) testicular neoplasms, none of which was stated to have been in a cryptorchid animal.

Schlotthauer, McDonald and Bollman [21] studied 82 spontaneous testicular tumors in dogs at operation and necropsy. Tumors were present in 59 testes of the 48 dogs examined. The average age of the dogs with the 51 interstitial cell tumors was 12 years. In 7 testes 18 interstitial cell tumors of multicentric origin were discovered. In 8 cases the interstitial cell tumor was associated with seminoma in the same testis. The average age of the 25 dogs with seminoma was over 10 years. Eighteen of the seminomas were of multicentric origin, and 4 were encountered in undescended testes. Of the 3 dogs with bilateral seminoma, 2 had scrotal and 1 had inguinal testes. Eighteen seminomas were in situ; 17 were not associated with other tumors and 5 occurred alone with interstitial cell tumors. These authors observed 6 testicular adenocarcinomas of varying degrees of malignancy.

Zuckerman and McKeown [4] examined histologically the testes and prostates of 243 dogs of various breeds. They found 35 testicular tumors: 17 seminomas in dogs 8 to 15 years old, 15 adenocarcinomas in dogs 4 to 15 years old and 3 interstitial tumors in dogs 9 to 15 years old. Innes [22] collected 49 canine testicular tumors, 36 of which were surgically removed. The ages of the animals, accurately tabulated by Innes, ranged from 5 to 13 years in 46 of the 49 cases. In addition to these 49 testicular neoplasms, including 32 seminomas, 15 tubular adenomas or Sertoli cell tumors and 2 cancerous interstitial cell tumors, Innes also saw nodular hyperplasia of the interstitial cells of the testes in 12 dogs. He presented evidence that nodular hyperplasia of the interstitial cells is not truly neoplastic. In his follow-up of surgical cases and from autopsies he concluded that the seminoma in the dog is of low grade malignancy as compared with the corresponding tumor in man. Of 79 growths of the testes of 393 dogs examined by Saloman,[23] 29 were seminomas, 41 were interstitial cell growths, 7 were mixed tumors and 2 were seminiferous adenomas, some of them multiple.

Although neoplasms of the testis comprise by far and away the greatest number of new growths observed in the canine male genital tract, several others of passing interest may also be mentioned briefly. Huebner [24] described the case of a male fox terrier, 2 years old, which had a bean-sized tumor of the dorsal surface of the penis and bilaterally enlarged inguinal lymph nodes. The dog was killed twenty-six days after the tumor had been extirpated, since it was blind. Autopsy disclosed metastases of small round cell alveolar sarcoma in the inguinal lymph nodes, in the right submaxillary lymph nodes and in the anterior chambers, irises and vitreous bodies of both eyes. These lesions were metastases from the primary sarcoma of the penis. Houdemer and Bablet [25] reported the case of a male mongrel dog, 18 months old, from which were surgically resected 2 nodules on the end of the penile sheath and abundant vegetations encircling the base of the penis and distending its sheath. The enlarged inguinal lymph nodes on the left side became infected and spontaneously obliterated, and those on the right side were resected twenty-four days later. The histologic diagnosis of the penile and lymph node lesions was lymphoblastoma. Little [26] saw a calcified squamous cell carcinoma of the penis of an old mongrel. Chambers [18] described a sheep dog, 10 years old, with a carcinoma of the prepuce.

Boucek [27] observed a dog with a fist-sized adenocarcinoma of the prostate and metastases in the pelvic and mesenteric lymph nodes. Described by Morini [28] was a wolf hound, about 2 years old, with an adenocarcinoma of the prostate which had extended widely to the pelvis and the pelvic viscera and metastasized to the kidneys, the liver and the mediastinal lymph nodes. Since the prostate also showed areas of adenoma, cystadenoma, fibromuscular hyperplasia and focal chronic inflammation, the author discussed the possibility of the change of a prostate involved in benign hypertrophy into one with an adenocarcinomatous character. Mori [29] reported the case of a great Dane, 8 years old, with a fist-

20. Cited by Krause.[8]

21. Schlotthauer, C. F.; McDonald, J. R., and Bollman, J. L.: J. Urol. **40**:539, 1938.

22. Innes, J. R. M.: J. Path. & Bact. **54**:485, 1942.

23. Saloman, cited by Innes.[22]

24. Huebner: Berl. tierärztl. Wchnschr. **38**:135, 1922.

25. Houdemer and Bablet: Bull. Soc. path. exot. **20**:344, 1927.

26. Little, G. W.: J. Am. Vet. M. A. **71**:171, 1927.

27. Boucek, Z.: Arch. f. wissensch. u. prakt. Tierh. **32**:585, 1906.

28. Morini, E.: Boll. d. Soc. Eustachiana **29**:185, 1931.

29. Cited by Morini.[28]

sized adenocarcinoma of the prostate and metastases in the testes, lymph nodes, the spleen and the lungs. Recorded by Boudet [29] was the case of a dog with a carcinoma of the prostate and a metastasis in the right cerebral hemisphere. Rudduck and Willis [30] described an Alsatian, 9 years old, with an adenocarcinoma of the prostate and metastases in lymph nodes, the lungs and a kidney.

NEOPLASMS OF THE FEMALE GENITAL TRACT

Exclusive of mammary tumors, the neoplasms of canine female genitalia recorded in the literature are relatively few. With respect to the gonads, the ovarian neoplasms reported are far behind testicular new growths. Bruckmüller [31] mentioned a carcinoma of a canine uterus with nodular extension into the uterine wall and an oviduct. In female dogs Boucek [27] saw a pedunculated fibromatous vaginal polyp, an ulcerated squamous cell carcinoma of the vagina and a walnut-sized leiomyoma of a gravid uterus. Jöhne [32] listed 1 uterine and 2 vaginal leiomyomas. Leisering [32] described a bitch with multiple submucous fibroleiomyoma of the vagina. In a 7 year old female brach hound Roquet [33] observed a pedunculated vaginal fibroma which hindered micturition by reason of its attachment in front of the urethral meatus. Belkin's [34] case 238 was that of a female Irish setter, 6 years old, with a submucous fibroleiomyoma on the posterior vaginal wall. Little [26] observed multiple neurosarcoma in the vagina of an English setter.

Coquot and Nenkoff [35] culled 18 ovarian neoplasms from 31 female dogs with abdominal tumors. Eight were ovarian and extraovarian cysts, 5 were cysts and adenomas, 3 were carcinomas, 1 was a lymphadenoma and 1 was a fibroma. The symptoms accompanying these ovarian tumors were increase in the size of the abdomen, panting, interference with locomotion and weakness and infrequency or absence of the estrual periods. The abdominal examination revealed a palpable, mobile solid mass which within limits could be grasped through the abdominal wall. The ovarian tumor was usually rounded, nodular, displaceable in the abdominal cavity by the thrust of the fingers, and was rendered immobile by distention of the edematous ligament attaching it to the lumbar gutter. Depicted in this paper were a 9 year old shepherd bitch with a 5,300 Gm. fibroma of the left ovary and an 8 year old hunting dog with cysts and adenomas of the right ovary weighing 6,600 Gm. Huggins and Moulder [36] mentioned a dog with carcinoma of the uterine cornua and 3 dogs with vaginal leiomyoma.

NEOPLASMS OF OTHER ENDOCRINE GLANDS

Not available for perusal was a paper on hypophysial tumors by Luksch [37] (1923). Other neoplasms occurring in the canine pituitary gland (aside from solitary or multiple cysts, which are rather common in dogs according to Stockard [38] and in my experience) were 2 adenomas of the anterior lobe, 1 recorded by Goodpasture [5] and the other by Joest, previously quoted.[39] White [40] described a male fox terrier, about 4 years old, which showed irritability, lethargy, polyuria, polydypsia and obesity six months before death. Autopsy disclosed small testes, a small prostate, a large thymus and a suprasellar craniopharyngioma which was about four times the size of a normal pituitary gland and invaded the midbrain, the third ventricle, the crura cerebri and the optic thalami. No trace of a pituitary gland was discovered. White also mentioned 3 other tumors of the canine pituitary gland, including an adenocarcinoma (Joest), a chromophobe adenoma with the syndrome of adiposogenital dystrophy (Hare) and an adenoma (Belmonte).

Blair [41] reported the case of a Boston bull terrier, about 12 years old, which suffered from obesity, asthma, chronic cough and dyspnea. At autopsy a globular mass was adherent to the superior borders of the cardiac auricles, was attached quite firmly to the trachea and the esophagus and surrounded the pulmonary veins. The heart was greatly hypertrophied, the ventricles were dilated, and the atrioventricular valves were marked by vegetation and imperfect closure. Although the diagnosis of "round cell sarcoma in the heart" was made, the obvious conclusion is that this neoplasm was a lymphosarcoma of the thymus. A thymoma reported by Joest has been mentioned before.[39] Goodpasture [5] described a "reticular" tumor of the thymus.

30. Rudduck, H. B., and Willis, R. A.: Am. J. Cancer **33**:205, 1938.
31. Bruckmüller, cited by Casper, M.: Ergebn. d. allg. Path. u. path. Anat. **3** (pt. 2):754, 1896.
32. Cited by Casper, M.: Ergebn. d. allg. Path. u. path. Anat. **11**(pt. 2):1068, 1907.
33. Roquet, M. M.: J. de méd. vét. et de zootech. **59**:713, 1908.
34. Belkin, G.: Berl. tierärztl. Wchnschr. **41**:829, 1925.
35. Coquot, A., and Nenkoff, G.: Rec. de méd. vét. **106**:129, 1930.
36. Huggins, C., and Moulder, P. V.: J. Exper. Med. **80**:441, 1944.
37. Luksch, cited by Winkler, K.: Ergebn. d. Biol. **5**:692, 1929.
38. Stockard, C. R., and others: The Genetic and Endocrine Basis for Differences in Form and Behavior, American Anatomical Memoir 19, Philadelphia, Wistar Institute of Anatomy and Biology, 1941, p. 454.
39. Mulligan, R. M.: Arch. Path. **38**:115, 1944.
40. White, E. G.: J. Path. & Bact. **47**:323, 1938.
41. Blair, W. R.: J. Am. Vet. M. A. **49**:520, 1916.

To my knowledge no one has ever recorded an instance of a solid neoplasm of the parathyroid glands of a dog. On the other hand, the thyroid gland is frequently the site of nodular hyperplasia [42] as well as of true neoplasm. Ewald [43] reviewed 75 cases of cancerous canine goiter from the literature and added 5 new cases of his own. In 63 of the 80 cases the neoplasm was a carcinoma, in 6 a sarcoma and in 7 a mixed tumor (carcinoma and sarcoma coexisting). In 4 it was not classified as to histologic type. The cervical veins were invaded in 6 cases, and metastases involved the cervical lymph nodes in 3 cases. The other organs with metastases and their frequency were as follows: lungs, 33; spleen, 9; kidneys, 6; mediastinal lymph nodes, 5; heart, 4; liver, 4; testes, 2, and skin, adrenal glands, breast, and intestine, 1 each. Reports of other instances of cancer of the thyroid gland have been abstracted in previous papers.[44] Another case was described by Cremona [45]; a male pointer, about 7 years old, was observed with a polymorphous sarcoma of the thyroid gland and metastases in the lungs, the left kidney and the bronchial, mediastinal and left lumbar lymph nodes. Rudduck and Willis [30] saw a male sheep dog, 10 years old, with a mixed adenocarcinomatous and ossifying neoplasm of the thyroid gland and ossifying metastases in the lungs.

Neoplasms of the pancreatic islets [46] have already been mentioned. Aside from the nodular hyperplasia of the adrenal cortex discussed before,[42] no neoplasms of this structure have been found recorded. The cancer of the medulla of the adrenal gland described by Goodpasture [5] was the only true neoplasm of this organ discovered in the literature. This tumor invaded the adrenal veins and the inferior vena cava and metastasized to the liver.

INFECTIOUS OR VENEREAL SARCOMA

An introduction to this phase of canine oncology could not more suitably be obtained than by summarizing the experience of Beebe and Ewing [47] with infectious sarcoma of dogs and their analysis of the work of Sticker with respect to this disease. Sticker secured specimens of infectious tumors of the penis and the vagina studied by Wehr, Geissler, Smith and Washbourn, Sanfelice and Bashford and submitted them with his own specimens to a large number of German pathologists, all of whom agreed on the diagnosis of round cell sarcoma. Supported by this diagnosis of recognized authorities on the structure of tumors, Sticker compared the general features of the disease with cancer in man and showed on these grounds that the growth must be accepted as a true cancer. The following statements constituted his proof:

1. Clinically, the other infectious venereal diseases of dogs resembling lymphosarcoma always cause prolonged inflammation and multiple nodules (merely enlarged lymph follicles), while inoculation of the vagina with the true tumor produces little, if any, inflammation, which soon subsides and is followed by a solitary tumor, or at best two to three tumors, arising in the subepithelial tissue.

2. Histologically, the tumor is entirely different from any known infectious granuloma, is not accompanied by any inflammatory reaction in the neighboring tissues and produces metastases, which develop from cells carried by the blood stream, without participation of the tissue cells.

3. Experimentally, living cells must be transferred in order to secure a growth, while tumor emulsion that has been finely comminuted or strained through a porcelain or paper filter is innocuous.

4. The spontaneous transference of the tumor is paralleled in the cases of "cancer à deux" and of carcinoma of the upper lip transferred from the lower lip in man.

Beebe and Ewing admitted that Sticker's argument presented a body of evidence strongly in favor of the opinion that infectious lymphosarcoma of dogs is a true tumor. Their own experience supported the views of Sticker, although they were unable then to distinguish satisfactorily between his follicular vaginitis and lymphosarcoma. Beebe and Ewing were forced to conclude that infectious lymphosarcoma of dogs is a true cancer on the basis of the work of others and of their own experiments with this disease. The incidence of venereal sarcoma varied with different authors. Auler and Wernicke [48] found 8 (1.4 per cent) tumors of this type among 585 canine tumors; Feldman [49] saw 7 (8.6 per cent) among 81 tumors in dogs; Stubbs and Furth [50] observed 5 (0.017 per cent) among 30,000 dogs observed in the animal clinic in Philadelphia; Vos [51] collected 5 (0.5 per cent) from among 1,100 Javanese dogs, and Kaalund-Jørgensen and

42. Mulligan, R. M.: Cancer Research **4**:505, 1944.
43. Ewald, O.: Ztschr. f. Krebsforsch. **15**:85, 1916.
44. Mulligan (footnotes 39 and 42).
45. Cremona, P.: Nuovo Ercolani **26**:365 and 388, 1921.
46. Slye and Wells.[11] Bru.[12]
47. Beebe, S. P., and Ewing, J. A.: J. M. Research **15**:209, 1906.
48. Auler, H., and Wernicke: Ztschr. f. Krebsforsch. **35**:1, 1931.
49. Feldman, W. H.: Neoplasms of Domesticated Animals, Philadelphia, W. B. Saunders Company, 1932, pp. 343-356.
50. Stubbs, E. L., and Furth, J.: Am. J. Path. **10**: 275, 1934.
51. Vos, J. J. T.: Geneesk. tijdschr. v. Nederl.-Indië **75**:263, 1935.

Thomsen [52] described 8 (0.6 per cent) among 1,400 dogs. Wong and K'ang [53] successfully treated 3 female dogs with infectious sarcoma of the vagina with radium. One bitch also had involvement of the inguinal lymph nodes, which subsided after radium therapy. A male dog with an infectious sarcoma of the penis was given several courses of radium treatment, following which stricture of the penile urethra developed, with resultant uremia and death. This paper contained a good photomicrograph (× 1,000) of the tumor in their case 2.

Since infectious or venereal sarcoma of the dog is obtained spontaneously through coitus and since cohabitation of the dog is conditioned by receptivity of the bitch for the male, it is logical to assume that female dogs were first afflicted with this disease, although where it might have come from before the female dog acquired it is a matter of conjecture. Because the time of estrus, or the period of receptivity of the bitch for the male, is associated with the highest possible natural estrogenic stimulation of the canine female genital tract, notably the vagina, in consequence of the hormonal elaboration of the ripe ovarian follicles, the future investigation of venereal sarcoma in the dog might well be directed along the lines of hormonal research.

MAMMARY NEOPLASMS

Neoplasms of the mammary glands are probably as common as any occurring in dogs. Gibbes [54] saw an alveolar cell sarcoma in the mammary gland of a bitch. Kitt [55] described fibroma of the breast, seen frequently in dogs, as a sharply delimited nodular hard structure derived from the interstitial connective tissue and surrounding the mammary ducts and acini (fibroma pericanaliculare). He also mentioned myxofibroma of the canine breast. He stated that in the dog enchondroma is found chiefly in the breast, is often multiple and usually appears in association with mixed tumors. In the breasts of bitches he also observed osteoma and chondroosteoma. In most instances they were sharply circumscribed, round and nodular, but they also occurred as knotty flat bony bars.

Stockfleth-Bang [32] described a lipoma in the breast of a fat bitch and said that in the breast lipoma may reach a tremendous size. Lucet [32] reported a 1,700 Gm. myxoma of the right third breast of a 6 year old female sheep dog. Stenzel [32] described a 2,000 Gm. mammary chondroma in a dog. In this tumor he found many areas in which embryonal cartilage tissue was in various stages of development. He also reported 2 cases of cystic fibrochondroma of the canine breast. Grischin [32] removed from the breast of a bitch a mobile hard osteochondroma the size of a hen's egg. Petit [32] observed 2 mammary tumors, a pure chondroma and an osteochondroma, in the same breast of a dog.

Freese [56] was the first to review rather extensively the early literature on canine mammary neoplasms. He mentioned some cases already cited by Çasper [32] and many others. He also reported 7 cases of his own. In the early literature he found reports of several varieties of canine mammary tumors, including fibroma or fibroadenoma (McFadyean; Ball and Leblanc; Jöhne), myxoma (McFadyean), chondroma (Peuch; McFadyean; Fröhner), fibromyxochondroma (Jöhne), sarcoma (Peuch; Möller; Jöhne; McFadyean), sarcoma with widespread metastases (Petropawlowsky), chondrosarcoma (Jöhne and Fröhner), cystadenoma and adenocarcinoma (Jöhne) and carcinoma (Semmer; Jöhne; Casper; Fröhner). Putz [57] saw a dog with multiple mammary carcinoma and metastases in the lungs and the left elbow joint. Petit [57] described an old bitch with a mammary carcinoma and metastases in the liver, the spleen, the lungs and other organs. Kitt [57] observed a female dachshund with a mammary cystocarcinoma and metastases in the lungs. Stenzel [57] found an adenocarcinoma of the type of embryonic mammary anlages in the breast of a bitch.

Freese [56] studied both grossly and microscopically 7 canine mammary neoplasms which may be listed as follows: (1) a cystochondrofibrosarcoma of the last four breasts of a spitz 11 years old; (2) a cystadenosarcofibroma of the left penultimate breast of a mongrel spitz 16 years old; (3) an angiofibrosarcoma of the left hindmost breast and an osteochondrofibrosarcoma of the left penultimate breast of a dachshund 11 years old; (4) a cystadenofibrosarcoma of the left penultimate breast of an English greyhound 4 years old; (5) a myxofibrosarcoma of the right penultimate breast and a cystofibroadenoma of the right hindmost breast of a dachshund 13 years old; (6) a papillary cystadenoma of the left penultimate breast of a cart dog 5 years old; (7) a papillary cystadenoma of the right caudal breast of a St. Bernard 13 years old. No

52. Kaalund-Jørgensen, O., and Thomsen, A. S.: Ztschr. f. Krebsforsch. **45**:385, 1937.
53. Wong, A. I. H., and K'ang, H. J.: Chinese M. J. **46**:377, 1932.
54. Gibbes, cited by Sutton, J. B.: J. Anat. & Physiol. **19**:415, 1884-1885.
55. Kitt, cited by Casper, M.: Ergebn. d. allg. Path. u. path. Anat. **3**(pt. 1):692, 1896.
56. Freese, K.: Ztschr. f. Tiermed. **9**:206, 1905.
57. Cited by Freese.[56]

metastases were seen at autopsy in any of the dogs. In the first 5 tumors pericanalicular tissue was most prominent; in the last 2 glandular tissue was paramount. According to Freese,[56] there occurs in the breasts of dogs a group of tumors with stroma composed of mesenchymal tissue that was broken off in embryonic life and subsequently began to grow. This mesenchymal tissue is undifferentiated and is variously ready to produce cells of connective tissue origin. Even more certainly, the epithelial structures of these tumors are derived from ectodermal sources and lie together with the mesenchymal elements in a resting state until, later, they begin to grow. These tumors are sharply delimited from the adjacent tissue and may be recognized as foreign bodies. These encapsulated mammary tumors are without exception mixed tumors, formed from various types of connective tissue elements and epithelial structures. They are usually benign and can be treated by operation. This discussion by Freese of the variegated structure seen in his own cases of mammary neoplasm as well as in those recorded in the early literature emphasizes their extremely complex structure and helps to explain many cases recorded since 1905.

Ortschild[58] recorded 5 mammary cancers as follows: (1) a cystadenocarcinoma of the right hindmost breast with metastases in the regional lymph nodes of a nulliparous Skye terrier bitch 16 years old; (2) an adenocarcinoma of the "second left thoracic breast" with metastases to axillary, thoracic and inguinal lymph nodes of an old Skye terrier bitch; (3) a cystadenocarcinoma of the third right breast with metastases in the opposite breast and the second breasts of an old bitch; (4) an adenocarcinoma of the right hindmost breast of a Gordon setter bitch 8 years old; (5) a papillary carcinoma of the right hindmost breast with metastases in inguinal lymph nodes of a Gordon setter bitch 10 years old. Foreman[59] reported the case of an Irish terrier bitch 11 years old from whose hindmost breast a 1 pound (0.5 Kg.) osteochondroma was extirpated. Seven months later autopsy disclosed metastatic osteochondroma in both the liver and the spleen. Goodpasture[5] saw 2 carcinomas of the female canine breast among 13 cancers culled from 50 old dogs.

Corsy and Thomas[60] found in a series of 50 canine mammary tumors, surgically removed, a liposarcoma occupying an entire breast. They also discussed 9 cases of human liposarcoma recorded in the literature. Among over 100 tumors of the canine breast surgically removed at the Alfort Veterinary School, Petit and Peyron[61] found a mammary tumor in which coexisted a predominantly epithelial mixed tumor and a liposarcoma.

Little[26] tabulated 12 canine neoplasms, among which were 5 located in the mammae of bitches as follows: (1) an adenocarcinoma of the left most caudal mamma of a Boston bull 9 to 10 years old; (2) a low grade mammary adenocarcinoma in a poodle; (3) an adenocarcinoma involving the first two right breasts and the left first breast, with metastases in the lungs and the spleen of a Boston terrier 13½ years old; (4) a mammary carcinoma in a fox terrier, and (5) a papillary adenochondrocarcinoma of the breast of a spaniel. Chambers[18] observed 15 histologically confirmed cases of canine cancer, among which was a recurrent mammary fibrosarcoma in an 11 year old fox terrier bitch. Rudduck and Willis[30] saw a female cocker spaniel with a cystic papillary mammary carcinoma.

Baldoni[2] reviewed cases of tumor of the canine male breast reported by Vachetta, Petit and Cinotti. Vachetta's case was one of spindle cell sarcoma. Petit described a male dog with a sarcocarcinoma of the breast and sarcomatous, carcinomatous and sarcocarcinomatous metastases in the lymph nodes and the lungs. Cinotti recorded the case of a male pug, 13 years old, with a mixed tumor (cystadenochondroma) of the right first abdominal breast. In addition to these Fröhner reported 1 and Murray 2 neoplasms of the canine male breast, which have been referred to in a previous review.[39]

The most common locations for mammary tumors in the dog are the posterior breasts. Of 114 mammary tumors studied and definitely located by Antoine, Liégeois and Verstraete,[62] 5 were situated in the first breasts, 18 in the second, 13 in the third, 36 in the fourth and 42 in the fifth. In 31 female dogs Huggins and Moulder[36] found 120 mammary growths of appreciable size, varying from solitary cysts to large solid tumors. These tumors were distributed in the breast as follows: first, 9; second, 18; third, 21; fourth, 36, and fifth, 36. The histologic types observed were sarcomatous, carcinomatous and papillary cystic. Metastases, consisting of intracystic papillary epithelial

58. Ortschild, J. F.: Bull. Johns Hopkins Hosp. **16**: 186, 1905.

59. Foreman, R. J.: Vet. J. **69**:240, 1914.

60. Corsy and Thomas: Bull. Assoc. franç. p. l'étude du cancer **16**:143, 1927.

61. Petit and Peyron: Bull. Assoc. franç. p. l'étude du cancer **16**:510, 1927.

62. Antoine, G.; Liégeois, F., and Verstraete: Bull. Acad. roy. de méd. de Belgique **14**:301, 1934.

tumors, were found in the lungs of 3 dogs and in the lungs and liver of a fourth.

In the experience of DeVita,[10] the growth of most mammary tumors in female dogs apparently parallels the growth of the endometrium during metestrus, their growth beginning possibly during estrus. They seem to be activated into growth at this time and then go into a quiescent phase during which they may become sclerotic. At the next estrus, growth seems to be reestablished, and they either increase in size or undergo regressive change, which in some instances results in complete destruction of the tumor through sloughing. The irregular growth may extend over several cycles. In 10 of 11 instances of mammary adenomatosis in which he also examined the uterus, DeVita observed atypical endometrial hyperplasia; this supports the assumption that estrogenic stimulation might be responsible for both the mammary and the uterine changes.

In considering mammary carcinoma in dogs from the point of view of etiology Little [26] said:

> . . . There is some evidence that a disturbance in the functional activity of the mammary glands, such as improper drainage of the milk flow resulting in mastitis, bears a causative relation to the late development of mammary carcinomas. This is frequently noticed in middle-aged or old virgin dogs, where a milk flow appears approximately 2 months after the oestrual period—a phenomenon occurring in non-pregnant bitches which has never been explained. Sometimes this lactation persists for 6 weeks to 2 months.

In their experiments with commercial betanaphthylamine, with which they produced tumor of the bladder in female dogs, Hueper, Wiley and Wolfe [63] observed swollen and lactating mammary glands in many dogs on various occasions during the latter third of the experimental period of over two years. The posterior mammae were most often affected. Whether the carcinogen was directly or indirectly responsible for this change was not elucidated.

On the basis of these data and of those presented in two other papers [44] it may be said that mammary neoplasms are among the most common tumors in dogs, occur most frequently in dogs 6 years of age or older, are obviously heavily preponderant in female dogs although they do occur in male dogs, may often be multiple, vary greatly in size and are most commonly located in the posterior breasts. The mixed tumor character of many of these growths is revealed in the varying combinations of cancerous and noncancerous changes in the cystic, solid and papillary cystic epithelial proliferations and in the growth of connective tissue, cartilage, bone, myxomatous tissue and even of adipose tissue. The types of tumors described include fibroma, fibroadenoma, osteoma, chondroma, osteochondroma, lipoma, myxoma, adenoma, cystadenoma, carcinoma, adenocarcinoma, cystadenocarcinoma, sarcoma, fibrosarcoma, chondrosarcoma, osteosarcoma, liposarcoma and myxosarcoma as well as varying combinaitons of these both cancerous and noncancerous neoplasms. Metastases of canine mammary cancer may be carcinomatous, sarcomatous or both. From the point of view of etiology two ideas have been advanced concerning the canine mammary neoplasms. The first is that they may be the result of congenital cell rests taking on abnormal growth characteristics. The second is that estrogenic stimulation of the mammary gland over several cycles may be responsible for their production. In the light of newer concepts of carcinogenesis it may appear that these two factors are but predisposing for one or more other factors responsible for actual neoplasia and even anaplasia.

63. Hueper, W. C.; Wiley, F. H., and Wolfe, H. D.: J. Indust. Hyg. & Toxicol. **20**:46, 1938.

SUMMARY

A review of the literature on canine oncology revealed several leads which might be followed in future investigation of neoplasms of the endocrine glands and of the genital tracts of both sexes. These included a syndrome of feminization in male dogs associated with testicular carcinoma, the relation between benign prostatic hypertrophy and castration, the coexistence of changes in the female genital tract with cysts and solid tumors of the ovary, the presence of hyperinsulinism with neoplasms of the pancreatic islets, the problem of the interpretation of proliferation of interstitial cells of the testes, the paucity of neoplasms reported for the pituitary, parathyroid, thymus and adrenal glands, the possible etiologic connection of hormones with venereal sarcoma, and the embryologic and hormonologic aspects of mammary tumors.

A THEORY OF TRANSPOSITION OF THE ARTERIAL TRUNKS BASED ON THE PHYLOGENETIC AND ONTOGENETIC DEVELOPMENT OF THE HEART

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The nature of the abnormality in transposition of the large arteries has intrigued pathologists for many years. The various theories that have been propounded have been reviewed elsewhere.[1] We have been engaged in studying this problem since 1935. In 1937 we presented a possible explanation of the anomaly.[1a] Since then we have enlarged our experience and have somewhat altered our original theory. We wish to present here our present conception of this anomaly based on our present knowledge of the phylogenetic and ontogenetic development of the heart.

THE ONTOGENETIC AND PHYLOGENETIC DEVELOPMENT OF THE HEART

Transposition of the large arteries cannot be understood without a knowledge of the evolution of the mammalian heart. This in turn is dependent on a knowledge of the evolution of the circulation of the vertebrates and the evolutionary progress of life as it emerged from sea water to fresh water and from fresh water to land. Already in the higher invertebrates (Annelida) a closed circulation is present. As the invertebrates changed their habitat from sea water to fresh water, the new environment led to the development of the vertebrates. Some of the vertebrates then migrated to land, while most others went back to the sea. The further evolutionary progress of the vertebrates was concerned with those that took up terrestrial life.

The outstanding characteristics of the terrestrial habitat are: (1) the presence of a relatively tremendous amount of oxygen in the atmosphere, (2) the low viscosity of the air, permitting rapid motion, (3) the marked fluctuations in the temperature and (4) the relative absence of water. The evolutionary progress of animal life on land is correlated with the increasing ability of vertebrates to utilize this environment and overcome its obstacles. Already in fresh water osmotic independence had been achieved. To survive on land, further adaptations were necessary, such as changes in the metabolism of nitrogen and adjustments for conservation of water and utilization of oxygen and finally for control of heat. The development of the circulation, although correlated with all these adaptations, was mostly related to the increasing ability of the organism to utilize oxygen.

The change from a water to a land environment necessitated a change from the gill type of respiratory apparatus. With the great increase in available oxygen, lungs were developed with a gradually increasing amount of respiratory epithelium. The increased absorption of oxygen and the increased liberation of carbon dioxide necessitated fundamental changes in the transport of these substances and in the giving of the one to, and the removal of the other from, the body cells. In other words, correlated with the tremendous increase in the oxygen content of the environment and the resultant gradual evolutionary changes in the ability to take in this substance, there ensued a gradual concomitant adaptation in the efficiency of the circulation. This adaptation was made by (1) an increase in the efficiency of the pump, (2) an increase in the oxygen capacity of the circulating medium and (3) an increase in capacity for oxygen exchange between the blood and the tissues.

The increased efficiency of the pumping system was obtained by (1) a gradual increase in pressure (Redfield [2]), (2) an increase in rate (Redfield [2]) and (3) a gradual separation of arterial and venous blood. Our task, therefore, is to analyze the morphologic changes in the heart

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1. (*a*) Lev, M., and Saphir, O.: J. Tech. Methods **17**:126, 1937. (*b*) Harris, J. S., and Farber, S.: Arch. Path. **28**:427, 1939.

2. Redfield, A. C: Quart. Rev. Biol. **8**:31, 1933.

producing this increased efficiency of the pumping system. Because of our problem of transposition a more detailed study must be made of the adaptational changes in the bulbus in the evolutionary series.

Beginning with the elasmobranch, the heart is more or less an undivided tubular heart consisting of a sinus venosus, a single auricle, a single ventricle and a bulbus leading into a common arterial trunk.[3] Blood from the arterial trunk is fed to the various gill arches for oxygenation and is then sent through the systemic circulation to the body cells. Unoxygenated blood is then returned to the heart by the venous circulation (fig. 1). In this type of uncomplicated circulation the bulbus, which is the lowest type of bulbus seen in the vertebrates, may be considered to have the following functions: (1) to prevent reflux of the blood into the ventricle, (2) to aid in propulsion of the blood and (3) to safeguard the fragile capillaries of the gills from excessive pressures (Keith[4]). In this species it is a comparatively long straight muscular tube in which the cavity is divided by four longitudinal ridges (anterior, posterior, right and left) extending throughout its length (fig. 2). These ridges are jammed together during cardiac contraction, thus preventing regurgitation into the ventricle (Robertson[5]). In the ganoid fish a similar undivided tubular heart and straight bulbus is present. However, instead of ridges in the bulbus, there are eight long rows of pocket valves, four of them (anterior, posterior, right and left) being more prominent than the others.

Thus in these early vertebrate forms both the internal respiratory (systemic) and the external respiratory (gill) circulation are controlled by one pump. This conforms to the relatively low amount of oxygen and carbon dioxide exchange in these forms. The bulbus here partakes in the propulsion of blood in addition to carrying on its valvular and gill-protecting functions.

A radical change in the circulation occurred in the dipnoan fish. This fish achieved air breathing in part and developed lungs. The increase in respiratory epithelium and the concomitant increase in oxygen and carbon dioxide exchange were accompanied by an increase in the size of the internal respiratory (pulmonary) circulation. This in turn was accompanied by a division of the pump system. The first stage of this is seen in the formation of the multiaxial auriculoventriculobulbar loop and the bayonet-shaped bulbus. By this method a relatively larger pumping power was obtained to cope with the necessity of increased pulmonary flow. This formation was correlated with the beginning formation of septums in the auricles and the ventricles, the formation of a bayonet-shaped bulbus containing a spiral valve and a twisting of the circulation of 180 degrees. This twist made possible an interchange of blood between the as yet only slightly divided circulations. The possible hydrodynamic explanations for these changes have been offered by Spitzer[6] and will be discussed later.

The changes in the bulbus of the dipnoan fish merit further attention. These have been described by Robertson[5] (fig. 2). The bulbus of the dipnoan fish, in contrast to the straight muscular tubular bulbus of the ganoid fish, is kinked in bayonet form, with a thinning out of the musculature of the ventral and lateral walls of the middle segment and the disappearance of the valves on these walls. Also in other segments of the bulbus the valvular structures have become smaller except at the distal end of the distal part of the bulbus, where they are still large and apparently functioning. In addition, a spiral septum with a twist of 270 degrees has been formed, going from right cushion 1 distally to left cushion A proximally. This extends from the middle of the distal to the proximal end of the proximal part of the bulbus. In the lepidosiren this ridge extends throughout the whole course of the bulbus, from the distal to the proximal end. In addition there is now a left ridge 3 in the distal bulbus. In both the ceratodus and the lepidosiren the proximal segment of the bulbus has a thick muscular coat, while the musculature of the distal and middle segments is relatively thinned.

These facts show that already in the dipnoan fish the muscular power of the bulbus was reduced, the muscle power being vested mostly in the ventricle, and the valve function in the bulbus had begun to be concentrated in the bulbus-truncus junction. In addition the bulbus was subjected to the process of septation just as were the other chambers of the heart. Thus the heart of the dipnoan fish was adapted to a greater intake of oxygen by an increase in pumping power and a beginning separation of the circulations. In the Amphibia the circulation was not much altered from that in the Dipnoi. The auricles are more completely separated, but there is still a common ventricle and the bulbus possesses the characteristics of the dipnoan bulbus.

3. Already in the lowest craniates the heart tube is not straight but is kinked on itself in one plane. This kink differs in complexity from the multiaxial loop of the dipnoan and higher vertebrates.

4. Keith, A.: Lancet **2**:1267, 1924.

5. Robertson, J. I.: J. Path. & Bact. **18**:191, 1913.

6. Spitzer, A.: Virchows Arch. f. path. Anat. **243**: 81, 1923.

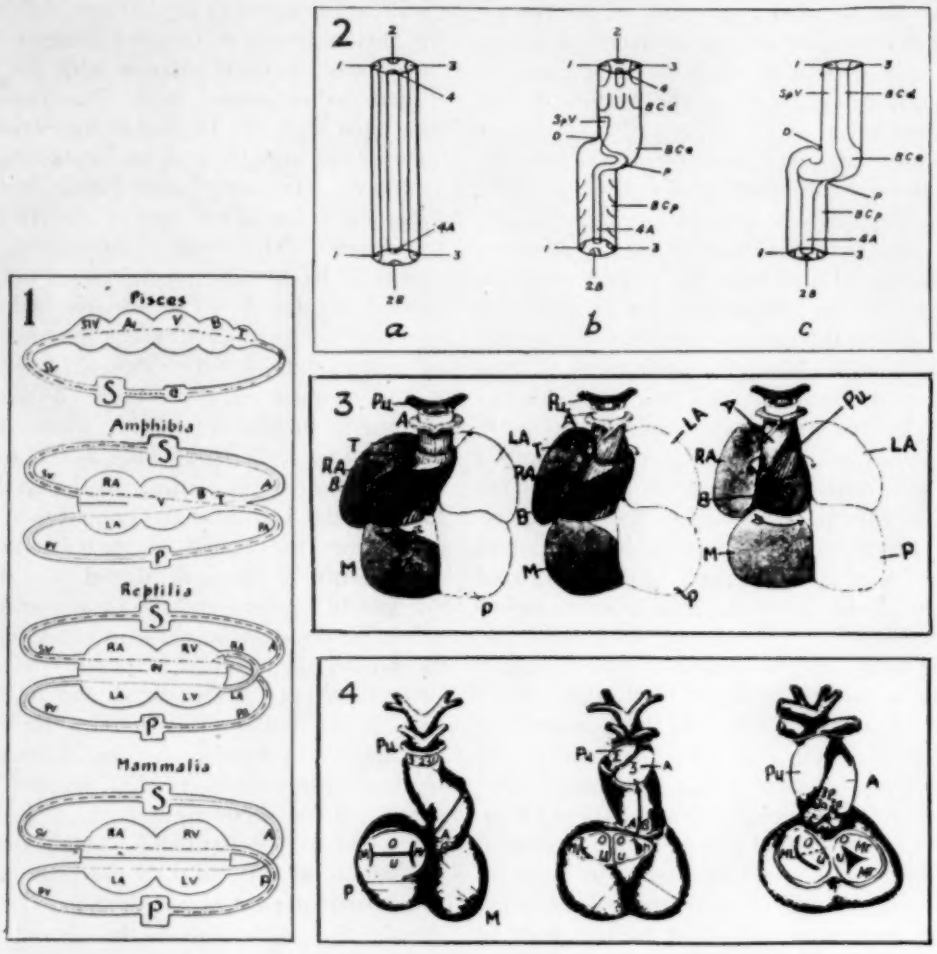


Fig. 1.—Diagrammatic sketches of the circulation in the phylogenetic tree. Pisces: The heart is a single tubular structure, consisting of a sinus venosus, an auricle, a ventricle and a bulbus (*SiV*, A_1, *V* and *B* in the diagram). Unaerated blood is pumped from the heart through the truncus (*T*) to the gills (*G*). The now aerated blood goes to the systemic cells (*S*). From there the venous blood is returned (*sv*) to the heart.

Amphibia: The auricular septum is now developed as well as the spiral valve (or partial septum) of the bulbus. The aortic portion of the bulbus is now decreased in size. These changes coincide with the appearance of the pulmonary vein and artery (*PV* and *PA*). Aerated blood in the left auricle (*LA*) and unaerated blood in the right auricle (*RA*) enter the common ventricle (*V*), where considerable mixing occurs. Mixed blood is then propelled from the ventricle into the bulbus (*B*), and from the bulbus it is propelled spirally through the truncus (*T*) into the systemic and pulmonary circuits (*A* and *PA*) from which it is returned (*sv* and *pv*) to the heart. *P* indicates the lungs.

Reptilia: A complete auricular and a partial ventricular septum has now formed (*DV* indicates the defect of the interventricular septum). The bulbus has almost completely disappeared. A right ventricular (left) and a left ventricular (right) aorta (*RA* and *LA*) are present. Unaerated blood is thus pumped from the right ventricle into the right ventricular (left) aorta and the pulmonary artery. Blood from the left ventricle is pumped into the left ventricular (right) aorta.

Mammalia: The auricular and ventricular septums are completely formed. The bulbus is completely absorbed. There is now complete separation of the systemic and the pulmonary circulation in the heart.

Fig. 2.—Diagrams of the bulbi of the hearts of (*a*) Elasmobranchii, (*b*) Ceratodus and (*c*) Lepidosiren paradoxa (from Robertson [5]). Note the formation of the spiral fold from right-sided ridge *1* in the distal portion, and the ventral ridge *4 A* in the proximal portion. *1*, right bulbar ridge. *2*, distal, and *2B*, proximal, part of the dorsal bulbar ridge. *3*, left bulbar ridge. *4*, distal, and *4A*, proximal, part of ventral bulbar ridge. *BCd*, distal segment of the bulbus. *BCp*, proximal segment of the bulbus. *BCe*, transverse segment of the bulbus. *D*, distal constriction of the bulbus. *P*, proximal constriction of bulbus. *SpV*, spiral valve.

Fig. 3.—Second phase of the development of the heart—the absorption of the bulbus (from Pernkopf and Wirtinger [7]). *LA*, left auricle. *RA*, right auricle. *P*, proampule. *M*, meta-ampule. *B*, bulbus. *T*, truncus. *Pu*, pulmonary artery. *A*, aorta.

Fig. 4.—Second phase of the development of the heart—changes in the auricular canal (from Pernkopf and Wirtinger [7]). *O*, *U*, *ML*, *MR*, endocardial cushions. *A*, *B*, proximal bulbar cushions. *1*, *2*, *3*, *4*, distal bulbar cushions. *Pu*, pulmonary artery. *A*, aorta. *M*, meta-ampule. *P*, proampule.

In the reptile further modification of the circulation occurred, producing a more complete separation of the pulmonary and systemic circulations and an increase in the efficiency of each. The auricular septum is now completed and only a defect remains in the ventricular septum (except in the crocodile, in which it is complete). The bulbus now underwent still more marked changes from that present in the Amphibia, changes directed toward its almost complete absorption into the ventricles, with the transmission of its functions to the ventricles and the truncus. Thus in Lacerta during embryologic development the bulbus becomes shortened and straightened with the disappearance of the distal and middle segments. The proximal portion is absorbed into the ventricles, all except a comparatively narrow rim of muscle at the base of the great arterial trunks (Robertson [6]). The bases of the great vessels are now guarded by semilunar valves. Thus, in addition to a diminution in the muscular power of the bulbus which had already occurred in the Dipnoi, the actual absorption of the bulbus and its elimination as a cardiac chamber must have been begun in the primeval reptiles and has proceeded far in present day reptiles. The functions of this chamber were taken over by other parts of the heart tube. Its valve function, to prevent regurgitation of blood into the ventricles, was taken over by the semilunar valves. Most of its musculature disappeared as contractile power was concentrated in the ventricles. And, according to Keith,[4] the portion of the bulbus which remained as the infundibulum of the right ventricle retained its function of shock absorber to the pulmonary circulation.

Pernkopf and Wirtinger [7] have analyzed the process of the absorption of the bulbus as it occurs in the reptiles. It may be subdivided into two components. First to appear is torsion at the ostium bulbotruncare in a counter-clockwise direction (if one is looking bulbusward from the ventricles). Then this becomes augmented in higher reptiles by back torsion (clockwise if one is looking bulbusward from the ventricle) at the ventriculobulbar ostium. By these processes the twist of the bulbar ridges is almost completely eliminated and the bulbus is absorbed into the ventricles. The first process (torsion at the ostium bulbotruncare) occurs maximally in lower reptiles—Lacerta and land turtles, in which it is greater than in mammals but in which ventral deviation and back torsion of the proximal bulbus are minimal. In the sea turtles a moderate amount of each is present. In the crocodiles there is less torsion at the distal bulbar ostium with more back torsion at the proximal bulbar ostium. Thus apparently there is an inverse relation between torsion at the distal bulbar ostium and back torsion and ventral deviation at the proximal ostium. Both of these processes result in the elimination of the twist of the bulbar ridges, telescoping of the bulbus and its absorption into the ventricles. In addition back torsion and ventral deviation of the bulbus facilitate the completion of the ventricular septum. Thus the reptilian embryo goes through an additional process in the development of the heart. In addition to the formation of the auriculoventriculobulbar loop and the bulbar bayonet, with greater perfection of septation, it has now gone a step further in the absorption of the bulbus.

The completion of the task of separating the circulations with the development of greater efficiency in each occurred in the mammal. Here the embryo in its development, in addition to recapitulating the embryonic stages of its phylogenetic ancestors (Needham [8]), went a step further in the absorption of the bulbus, and completed the process of septation and the separation between the systemic and pulmonary circulations. Pernkopf and Wirtinger [7] have presented a very complete picture of this process.

The mammalian heart in its early development is an undivided tubular structure reminiscent of the elasmobranch and the ganoid fish, consisting of a sinus venosus, an auricle, a ventricle and a bulbus. The movements of the heart from its straight tubular form to its definitive condition proceeds in two phases. In the first phase (1 to 7 mm. embryo) the auriculoventriculobulbar loop and bulbar bayonet are formed. During these formations a torsion of 90 degree (clockwise, looking ventricleward from the auricle) occurs at the auriculoventricular region, and an opposite torsion of 90 degrees (counterclockwise, looking bulbusward from the ventricle) occurs at the bulboventricular region. These bring the mesocardial (dorsal) portion of the ventriculobulbar orifice to the left, and the mesocardial (dorsal) portion of the auricular canal to the right. At 4 to 5 mm. of fetal length the anlage of septums begins to be laid down in all portions of the heart tube (septum primum) as are the counter ridges of the auricle, the endocardial cushions, the main and the counter ridges of the ventricle, the proximal bulbar cushions, A and B, and the distal bulbar cushions, 1, 2, 3 and 4. These ridges are laid down on heterogonial (pertaining to the opposite anlage) parts of the heart tube sub-

7. Pernkopf, E., and Wirtinger, W.: Ztschr. f. Anat. u. Entwcklngsgesch. **100**:563, 1933.

8. Needham, J.: Biol. Rev. **5**:142, 1930.

sequent to the beginning and simultaneous with the later course of the development of the first phase of the movements of the heart. When first seen the auricular ridges have a negative torsion of 90 degrees, the ventricular ridges practically no torsion and the bulbar ridges a positive torsion of 270 degrees. It is thus evident that the formation of the multiaxial auriculoventriculobulbar loop and bulbar bayonet results in the laying down of a "heterogonial" septal anlage with resultant twisting of the as yet incompletely divided circulations by 180 degrees. By this method the future exchange of blood between the two circulations is effected. This process, which we have now outlined more fully, has already occurred, as mentioned, in the embryologic development of the Dipnoi and is present in all higher animals. It is correlated with the necessities arising from the change to air breathing and the appearance of lungs in the phylogenetic series.

The mammalian embryo proceeds now to its second phase of development (figs. 3 and 4). This phase concerns itself mainly with the absorption of the bulbus (fig. 3), a process that occurs only in amniotes (reptiles, birds and mammals) but, as mentioned, is incomplete in reptiles.[9] In mammals the absorption of the bulbus is effected by the two processes which we have outlined as they are seen in reptiles but in different degrees. Thus a torsion of 150 degrees (counterclockwise, looking truncusward from the bulbus) occurs at the ostium bulbotruncare, and back torsion of 45 degrees (clockwise, looking bulbusward from the ventricle) (first 90 degrees then reduced to 45 degrees) at the ostium ventriculobulbare. The latter is accompanied by a deviation directed ventrally and to the left of the bulbus. These movements are further correlated with a shift of the auricular canal to the right (fig. 4) with expansion of the tricuspid orifice and of the dorsal wall of the meta-ampule (distal portion of the primitive ventricle), shrinkage of that part of the proampule (proximal portion of the primitive ventricle) which borders on the cranial wall of the auricular canal, and shrinkage of the mesocardial wall of the proximal bulbus. In this way the bulbus is absorbed into the ventricles, its mesocardial (aortic) portion disappearing and its ventral portion assuming the role of conus of the right ventricle. The functions of the bulbus are now taken over by other parts of the heart. Its contractile function is taken over by the ventricles. Its valvular function of preventing backflow of blood is taken over by the semilunar valves, and its function of acting as a shock absorber for the pulmonary circulation is possibly taken over by the conus of the right ventricle (Keith [4]). The two circulations have now been completely separated by a completed ventricular as well as an auricular septum.

Thus the mammalian embryo has gone a step further than its reptilian ancestors. It has now reached what at present is the highest form in the evolutionary development of the heart—a complete separation of the circulations for internal and external respiration, correlated with the gradually increased ability to take in oxygen. (This is present also in the birds.) This ability is correlated with a gradual increase in blood pressure and rate in the vertebrate series culminating in the mammal (Redfield [2]), making possible the better transport of oxygen.

To understand more fully the transition from the reptilian to the mammalian stage of circulatory development (which is necessary for adequate interpretation of congenitally malformed hearts), a comparison of the bulbus and the truncus of the two species must be made. In the truncus, in both the crocodile and the mammal, the septum aorticopulmonale develops. Because of the presence of a fifth branchial arch in the reptile, this septum develops from the spurs between arches 4 and 5 in the reptile, while in the mammal it originates from the spurs between arches 4 and 6. In both species the fusion of these spurs and their caudal extension divide the truncus into a dorsal pulmonary portion and a ventral systemic portion. However, there is a difference in the relative size of the pulmonary portion as seen by the attachment of the septum aorticopulmonale to the distal bulbar cushions, 1 and 3. In the reptile the attachment is more toward the mesocardial end of these cushions, while in the mammal it is more toward the center of these cushions. In the reptile in addition to the septum aorticopulmonale a septum aorticum is formed. This originates from the spur between left arches 3 and 4, extends downward throughout the truncus, with its counterextension sinking into the right portion of the septum aorticopulmonale. This thus divides the anterior portion of the truncus into a small left portion consisting of the fourth left arch, and a larger right portion leading into both carotid arteries and the right fourth arch. The left portion connects with the right ventricular aorta, while the right portion connects with the left ventricular aorta. The horns of the septum aorticum reach on the left side the commissure between cushions 3 and 4

9. Pernkopf and Wirtinger failed to mention that a reduction in the musculature of the bulbus already occurs in amphibians and lung fish. Hence the second phase is really a further elimination of the bulbus rather than a separate process. However, we retain their terminology for convenience.

at ostium bulbotruncare and on the right side the anterior part of cushion 1. Thus in the reptile the truncus is divided into three vessels, two systemic and one relatively small pulmonic, while in the mammal it is divided into only two vessels, a systemic and a relatively larger (as compared with that of the reptile) pulmonic vessel.

The bulbus of the reptile and that of the mammal likewise show points of difference (fig. 5). In both there are four cushions, 1, 2, 3 and 4, at the distal ostium of the bulbus and two cushions, A and B, at the proximal ostium. In both in an early stage of development there is also cushion C, situated to the right between A and B. However, this cushion disappears before the septums form in the bulbus. The difference between the two species lies in the ridge connections between the proximal and distal bulbar cushions and the extent of their twist. In both, ridge 1A (homologous to the spiral valve of the Amphibia) is developed with a twist of 270 degrees. However, there are two further ridges in the reptile, 4B and a ridge going from cushion 3 to a point where cushion C originally was (we may call it 3C). In the mammal only a single opposite ridge 3B is developed. Neither of the ridges 4B and 3C has the twist of ridge 3B in the mammal. In the reptile are thus developed two septums in the bulbus—septum aorticopulmonale from 1A and 3C and septum aorticum from ridges 1A and 4B, each continuous with the similar septums in the truncus. In the mammal, however, a single septum is developed from ridges 1A and 3B. Thus are developed a right and a left aorta and a pulmonary artery in the reptile, and a single aorta and a pulmonary artery in the mammal.

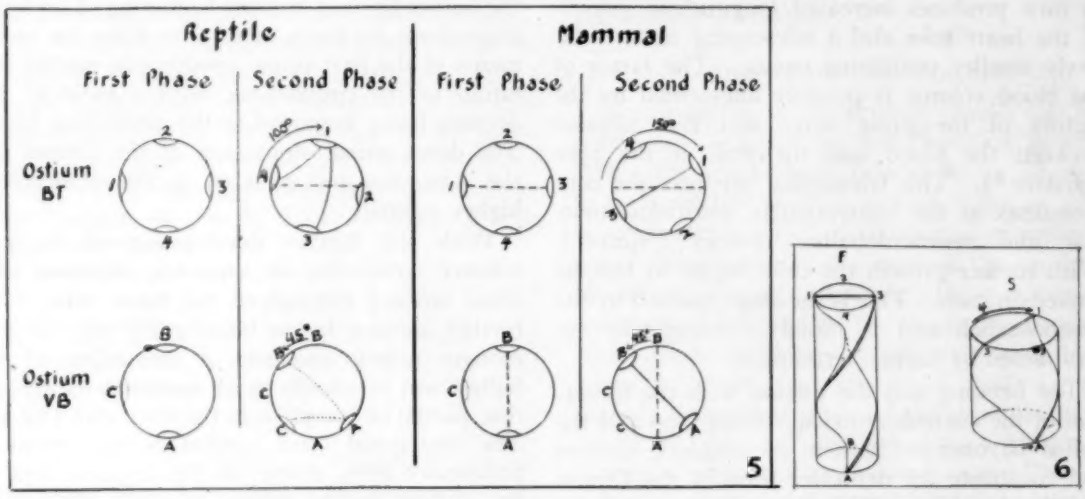


Fig. 5.—Comparison between the bulbus of the reptile and that of the mammal. Note that the number and the location of the bulbar cushions are the same. The difference lies in the number of ridges extending through the bulbus, connecting the distal and proximal cushions. There are three ridges in the reptile—ridges 1A, 4B and 3C. In the mammal there are only two ridges—1A and 3B. Therefore two septums are formed in the reptilian bulbus, but only one septum is formed in the mammalian bulbus.

Fig. 6.—Possible mechanical explanation for the absorption of the bulbus. *F*, first phase. *S*, second phase. *1, 2, 3, 4*, distal bulbar cushions. *A, B*, proximal bulbar cushions. *1A* and *3B*, bulbar ridges.

POSSIBLE HYDRODYNAMIC EXPLANATION FOR THE ONTOGENETIC AND PHYLOGENETIC DEVELOPMENT OF THE HEART

We have thus traced the phylogenetic and ontogenetic development of the vertebrate heart. The understanding of the possible forces involved in this development we owe for the most part to Spitzer.[6] Although Robertson[5] some years previously had pointed out the correlation between the changes in the bulbus and the development of the lungs, it was Spitzer who first suggested the possible hydrodynamic principles involved in the changes in the circulation in the phylogenetic series correlated with the development of the lungs. Utilizing the morphologic analysis made by Pernkopf and Wirtinger we have slightly extended Spitzer's conception. We present it in the following hypothesis, which is offered for the purpose of serving as an aid in the study of congenital anomalies.

The function of any circulation is the transportation and supply of materials for the metabolism of the component cells of the body and the removal of the waste products of metabolism. The differentiation of red blood cells during embryologic development and the simultaneous development of sinusoids and eventually of vascular tubing first to house and eventually to

transport the red blood cells thus may be correlated with the growth in number of the remainder of cells in the embryo and their further specialization necessitating and calling forth (by biologic forces as yet unknown) a circulation. Also, with greater specialization of body cells, the circulatory mechanism likewise becomes more specialized (by biologic forces as yet unknown), so that portions of the tubular structure take over the power of propulsion and hence the development in phylogeny and ontogeny of a straight tubular heart. When the circulation has reached a tubular stage, certain hydrodynamic principles may be assumed to act in its further development. The rapid increase in the number of systemic cells and the further specialization call forth a greater number of red blood cells and capillaries from the mesenchyme to house them. This increases the blood volume which in turn produces increased longitudinal growth of the heart tube and a telescoping in its relatively smaller containing cavity. The factor of the blood volume is possibly augmented by the factors of the pulse wave and the collision between the blood and the wall of the tube (Spitzer [6]). This telescoping produces the constructions at the sinoauricular, auriculoventricular and ventriculobulbar orifices (Spitzer). With further growth the tube begins to become kinked on itself. This is the stage reached by the elasmobranch and is rapidly telescopically recapitulated by higher vertebrates.

The bending and the torsion with the formation of the auriculoventriculobulbar loop and the bulbar bayonet are seen in the amniotic embryo and constitute the definitive form of the Dipnoi and the Amphibia. This formation is correlated with the development of the pulmonary circulation. The lungs represent a greater oxygen exchange mechanism than do the gills because of their relatively greater respiratory surface. This in turn is correlated with an increased capillary bed. The increase in number of "respiratory" capillaries is correlated with a marked increase in blood volume, and both are correlated in the phylogenetic development with the greater amount of oxygen in air as compared with sea and fresh water. The increase in blood volume produces a further increase in the size of the heart tube in comparison with its containing cavity, and thus it goes through the movements of the first phase with the accompanying torsions in multiple planes and the formation of a bayonet-like kinked bulbus. With the increase in blood flowing through the truncus into the pulmonary artery there are relative widening and shortening of the truncus. The centripetal force affects also the dividing spur between the fourth and sixth arches so that it is pulled toward the heart tube. At the same time the spur is stimulated to growth because of the irritation produced by the bilateral pressure of the increased volume. Thus we have beginning septation in the truncus. A similar process occurs at the venous end stimulating the growth of the spur between the two venous ostiums, leading to septation in the auricular regions. The same factors produce lateral distention and relative shortening of the dilated portions of the heart tube. The contracted portions between these dilated regions are thus pulled in the opposite direction with production of longitudinal folds. These are the distal and proximal bulbus swellings, the endocardial cushions and the sinoauricular folds. Thus terrestrial life is accompanied by the development of lungs, which is correlated with the development of the multiaxial auriculoventriculobulbar loop, the formation of the bulbar bayonet and the beginning of septation throughout the heart tube. These are the movements of the first phase resulting in partial septation of the circulations, with a twist of 180 degrees being imparted to the circulating blood. The development stops here in the Dipnoi and the Amphibia but goes on in the embryos of higher animals.

With still further development of the pulmonary circulation in amniotes, septation proceeds onward throughout the heart tube. The further increase in the blood going into the pulmonary artery produces a shortening of the bulbus and eventually in all amniotes its absorption, partial or complete, in the ventricles (fig. 6). The centripetal force created by the increased pulmonary flow, acting on the truncus, acts on the curved bulbar ridges as well, eliminating most of their twist and handing this to the truncus. The result is torsion at the ostium bulbotruncare. The same increased force acting from the arterial side acts on the venous side. With increased flow through the pulmonary veins, the hydrodynamic force acting in the opposite direction to that coming from the arterial side produces a back torsion and ventral deviation of the bulbus (fig. 3). The pulmonary flow in the reptile is less than that in the mammal, and this accounts for the morphologic differences in the bulbus and the truncus and in the absorption of the bulbus in the two species. The pulmonary portion of the truncus in the reptile is relatively smaller than that in the mammal. This is due to the more posterior attachment of the horns of the septum aorticopulmonale in the reptile as compared with the mammal, related to the smaller pulmonary flow. This permits the growth of the septum aorticum from the left caroticoaortic spur and brings about the adaptation of two aortas in present day reptiles.

In mammals the increased pulmonary flow, witnessed by the more anterior attachment of the septum aorticopulmonale to cushions 1 and 3, inhibits the formation of the septum aorticum. The bulbus of the mammal likewise shows the greater importance of the pulmonary circulation by the growth of two opposite ridges instead of three and the production of a single septum aorticopulmonale bulbi. The absorption of the bulbus of the mammal is more complete than that of the reptile, with the bulbar septum coming in line with the ventricular septum, completing the process of septation. The mesocardial portion of the bulbus, now being completely separated from the pulmonary portion, disappears or is telescoped into the left ventricle with complete absorption of the bulbus. The two circulations are now completely separated, and the efficiency of the pump has reached its highest point in the evolutionary development.

As mentioned earlier, in addition to the increase in efficiency of the pump, there was a second adaptation for the utilization of the high oxygen medium in the vertebrate series. This was the increase in the oxygen capacity of the blood. This was also a gradual evolutionary development, as seen in the accompanying table (Baldwin [10]):

	Cc. of Oxygen per 100 Cc. of Blood
Mammals	25.0
Birds	18.5
Reptiles	9.0
Amphibians	12.0
Fish	9.0

The increased amount of oxygen in the air, just as it must have served as the stimulus for changes in the lungs and in the pumping mechanism, must have served to increase the capacity of the blood for carrying oxygen. Hemoglobin is regularly present throughout the vertebrates and is the most efficient of oxygen-carrying pigments. Throughout the vertebrate series there is an increased morphologic differentiation of erythrocytes, coupled with an increased concentration of hemoglobin within them, accompanied by an increased number of them, all of which leads to greater oxygen carrying capacity (Redfield [2]). Because of the gradual increase of carbon dioxide produced by the cells in the vertebrate series, there ensued an increase of carbon dioxide–combining capacity and buffering capacity of the blood, which is seen in the evolutionary series accompanying the increase in hemoglobin. This property of carrying carbon dioxide, however, is not solely dependent on hemoglobin; it is correlated with serum albumin, globulin and certain inorganic substances, which are dependent on processes in the kidneys and other organs (Redfield [2]).

An accompaniment of the increase in oxygen capacity of the blood was an adaptation for more rapid and complete exchange of oxygen between blood and tissue—increased oxygen tension. This tension is low in fish and reptiles but high in birds and mammals (Redfield [2]).

Thus, in summary, the phylogenetic and ontogenetic development of the heart is part of the whole development of the circulation in the phylogenetic tree. This is mostly related to the change in habitat of the vertebrates, from fresh water to land. The interaction between the respiratory and circulatory system of these animals and the high oxygen content of the environment led to the progressive evolutionary development of the mammalian circulation.

A THEORY OF TRANSPOSITION

Transposition of the arterial trunks has been defined by Abbott [11] as that condition in which "the great trunks have undergone an alteration in their relative position to each other or to the ventricles from which they emerge, whereby the aorta comes to lie in the path of the unaerated blood from the right ventricle." Thus within this category of anomalies lie those conditions in which the aorta emerges from both ventricles or from the right ventricle alone, and the pulmonary artery springs from the right or the left ventricle. Also included are those conditions of mutually abnormally situated arterial vessels arising from a common or a slightly subdivided ventricle; likewise, truncus communis, truncus solitarius aorticus and truncus solitarius pulmonalis, in which the aortic vessel or component emerges from the right ventricle, or the aorta or its remnant is abnormally situated with respect to the pulmonary artery or its remnant.

Transposition of the arterial trunks may be classified as follows:

Type I (Spitzer). Riding aorta (Rokitansky)
(*a*) with an aneurysm of the membranous septum.
(*b*) with a defect of the ventricular septum.

Type II (Spitzer). Partial transposition (Rokitansky)—The aorta and the pulmonary artery arise from the right ventricle.

Type III (Spitzer). Complete transposition (Rokitansky)—The aorta comes from the right ventricle and the pulmonary artery from the left ventricle.

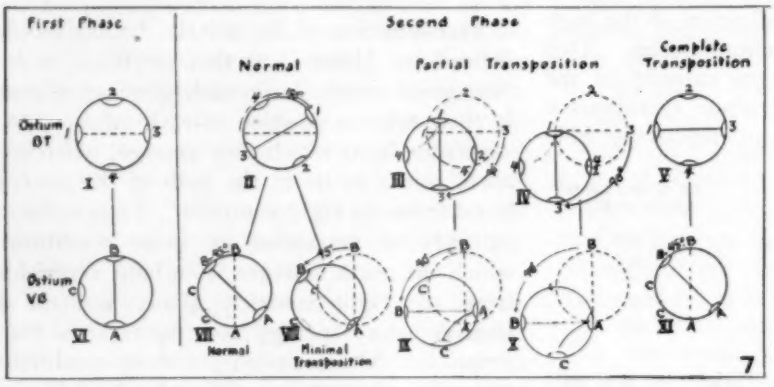
Miscellaneous group. Truncus communis, truncus solitarius aorticus with transposition, truncus solitarius pulmonalis with transposition, Type IV (Spitzer) and transposition with atresia of the tricuspid valve.

10. Baldwin, E.: An Introduction to Comparative Biochemistry, New York, The Macmillan Company, 1937.

11. Abbott, M. E., in Osler, W.: Modern Medicine, Phliadelphia, Lea & Febiger, 1927.

All types of transposition of the arterial trunks may be considered to be due to a "phylogenetic" abnormality complicated by further "ontogenetic" abnormalities as postulated by Spitzer.[6] The phylogenetic abnormality is in our opinion an abnormality in the formation of ridge 3 B. As pointed out, the normal mammalian bulbus presents two ridges, 1 A and 3 B, instead of the reptilian three ridges, 1 A, 4 B and 3 C (fig. 5). Ridge 1 A has already been developed in the lung fish (fig. 2). Ridge 3 B, however, is a new formation not present below the mammal. When in the embryo of our reptilian ancestor the first responses to a greater increase in oxygen intake began, thus starting the mammalian series, there apparently was elaborated a fusion of ridges 3 C and 4 B by the already described hydrodynamic principles. It was possible, with the aforementioned forces, for the bulbus to be completely absorbed, with production of the mammalian heart. The processes involved in this absorption of the most of the twist of the bulbar ridges and produced disappearance of the median bulbar segment, marked shortening of the bulbus, absorption of its ventral portion as the conus of the right ventricle and disappearance of the dorsal portion.

In the embryo whose parent form will present the anomaly transposition, although the stimulus is apparently there (as seen from the normal formation of lungs), it does not produce this new ridge 3 B but produces one or the other or neither of its components, 4 B and 3 C. In other words it is doing what most of the reptilian embryos did during the time our pioneering reptilian ancestor made a progressive change. Yet the hydrodynamic forces for the absorption of the bulbus are there The absorption of such a bulbus with a well formed ridge 1 A but an abnormal ridge 4 B and 3 C, or a poorly developed ridge 3 B, or with only ridge 1 A, proceeds abnormally (fig. 7). In the first place, the



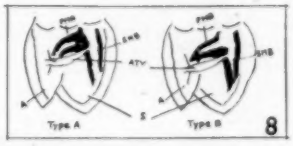


Fig. 7.—Normal and abnormal absorption of the bulbus. In transposition there is an abnormality in the formation of ridge 3B, whereby it is poorly formed or replaced by ridge 4B or 3C. This results in decreased torsion at the distal ostium. In addition, this torsion occurs either around cushion 1 as a center or close to cushion 1. At the proximal ostium back torsion occurs around cushion A as a center or close to cushion A. This back torsion may be 45 degrees, or it may be 90 degrees as is normally observed in an earlier stage, *1, 2, 3, 4*, distal bulbar cushions. *A, B, C*, proximal bulbar cushions. *BT*, ostium bulbotruncare. *VB*, ostium ventriculobulbare. *I, VI*, distal and proximal bulbar ostiums, respectively, at the end of the first phase (normal and in transposition). *II, VII*, distal and proximal bulbar ostiums during the second phase in normal absorption of the bulbus. *II, VIII*, bulbar ostiums in transposition with congenital aneurysm of the membranous septum during the absorption of the bulbus. *III, IX*, bulbar ostiums in partial transposition during the absorption of the bulbus when torsions occur close to cushions 1 and A as centers. *IV, X*, bulbar ostiums in partial transposition during the absorption of the bulbus when torsions occur about cushions 1 and A as centers. *V, XI*, bulbar ostiums in complete transposition in which no torsion occurs at the distal ostium while mild back torsion occurs at the proximal ostium.

Fig. 8.—Topography of the muscle bundles of the right ventricle. In *Type A* two distinct muscle bundles are noted. In *Type B* an arch of musculature is present at the base of the pulmonic valve. *SMB*, septal muscle bundle. *PMB*, parietal muscle bundle. *ATV*, anterior leaflet of tricuspid valve. *A*, anterior wall of the right ventricle. *S*, septal wall of the right ventricle.

bulbus were a torsion of 150 degrees (counterclockwise—looking truncusward from the bulbus) at the ostium bulbotruncare and a back torsion of 45 degrees (clockwise—looking bulbusward from the ventricle) at the ostium ventriculobulbare (fig. 3). The latter was combined with a deviation directed ventrally and to the left of the bulbus. These processes eliminated torsion at the distal and proximal ostiums cannot occur around the center of the tube as an axis; the center of rotation will be about a point between the center of the tube and cushions 1 and A, or on the periphery at cushion 1 and cushion A. In addition there will be less torsion at the distal ostium. For since we believe the torsion to be produced by the action of the centripetal

force on the spiral ridges, the presence of only one ridge with or without an opposite ridge with a lesser twist will lessen the torsion. The torsion of the proximal ostium may remain the same but will be eccentric with its center closer to A or at A. Thus the conuses of the aorta and the pulmonary artery will take up abnormal positions after the absorption of the bulbus. Hence the ontogentic complication of the phylogenetically abnormally formed bulbar ridges is the abnormal absorption of the bulbus.

TYPES OF TRANSPOSITION

We may now briefly discuss the various types of transposition and their underlying abnormalities.

associated with a mild degree of transposition. This was based on a study of the muscle bundles of the right ventricle in addition to the abnormal position of the aorta. In our opinion the underlying variant here is a poorly formed ridge 3 B resulting in a slight abnormality in the absorption of the bulbus. That is, normal torsion occurs at the distal ostium but slightly diminished back torsion occurs at the proximal ostium (*II* and *VIII* in fig. 7). This results in poor fusion of the proximal cushion of the bulbus, the endocardial cushions and the ventricular septum, leading to an aneurysm in the resultant pars membranacea.

Type I B—Riding Aorta with a Defect of the Ventricular Septum (fig. 9 *b*). Here the aorta, as in the previous type, straddles the muscular

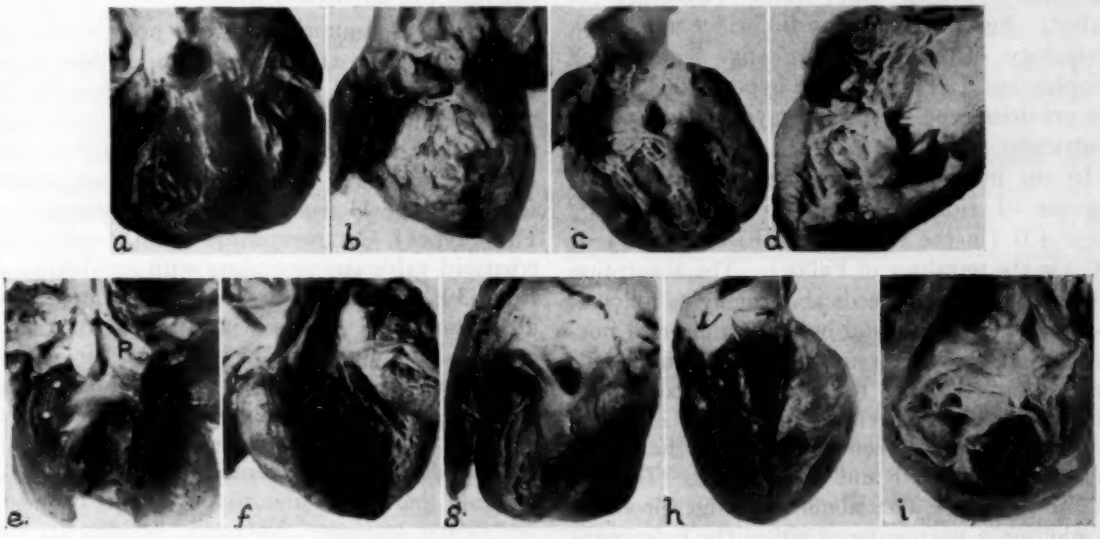


Fig. 9.—(*a*) Type 1A. This is a riding aorta with an aneurysm of the membranous septum; left ventricular view. (*b*) Type 1B. This is a riding aorta with a defect of the ventricular septum; right ventricular view. (*c*) Partial transposition with the tetralogy of Fallot; right ventricular view. (*d*) Partial transposition with the tetralogy of Eisenmenger; right ventricular view at the base of aorta. (*e*) Partial transposition with the tetralogy of Eisenmenger; right ventricular view at the base of the pulmonary artery (*P*, the pulmonary artery). (*f*) Complete transposition. (*g*) Truncus arteriosus communis persistens; right ventricular view. (*h*) Transposition with atresia of the tricuspid valve as it appears to one looking into the heart anteriorly. (*i*) Transposition with atresia of the tricuspid valve as it appears to one looking into the heart posteriorly.

Type I A—Riding Aorta with Aneurysm of the Membranous Septum (*fig. 9 a*).—In this type the aorta straddles the muscular ventricular septum over an aneurysm in the obliquely situated pars membranacea. The aorta is thus shifted slightly anteriorly, but the coronary arteries and ostiums show no evidence of transposition. More than 70 cases have been reported in the literature (Lev and Saphir [12]), in about 15 per cent of which the abnormal position of the aorta was noted. In our previous report on this anomaly we showed that congenital aneurysm of the membranous septum is often (if not always)

12. Lev, M., and Saphir, O.: Arch. Path. **25**:819, 1938.

ventricular septum, but instead of an aneurysm of the pars membranacea there is a defect of the ventricular septum. The aortic semilunar cusps are displaced in a slight counterclockwise direction (looking from the aorta toward the ventricle). The coronary arteries show an abnormal course typical of mild transposition (see Spitzer [6]). Stenosis of the pulmonary ostium with a bicuspid pulmonic valve may be present.

In our opinion the underlying abnormality here is a poorly formed ridge 3 B, this ridge being less in extent than in congenital aneurysm of the membranous septum. This results in abnormal absorption of the bulbus as follows: less than the normal 150 degrees of torsion in

the distal ostium and eccentric distal bulbar torsion and proximal bulbar back-torsion, as indicated in the prveious type (*III* and *IX* in fig. 7). Ridge 3 B does not meet the ventricular septum, thus preventing the completion of the latter, with the result that there is a defect in the anterior portion of the ventricular septum.

Type II—Partial Transposition (fig. 9 *c, d* and *e*).—In this type the aorta emerges from the right ventricle with the pulmonary artery. No vessel emerges from the left ventricle which presents the defect in the ventricular septum as its only outlet. The aortic semilunar cusps are still more rotated in a counterclockwise direction. The coronary arteries show a greater degree of transposition than in the previous type. The pulmonary orifice and trunk (tetralogy of Fallot) (fig. 9 *c*) or the aortic orifice and trunk (tetralogy of Eisenmenger) (fig. 9 *d* and *e*) (Saphir and Lev [13]) may be hypoplastic. As in the previous type there is always a defect of the ventricular septum.

In our interpretation the abnormality here is absence of ridge 3 B and its replacement by ridge 4 B (in the tetralogy of Eisenmenger) or 3 C (in the tetralogy of Fallot). The absorption of such a bulbus proceeds abnormally as follows: less torsion at the distal bulbar ostium and normal or 90 degree torsion at the proximal bulbar ostium. These torsions may occur with cushions 1 and A as centers (*IV* and *X* in fig. 7) or with a point between the center of the bulbar tube and cushions 1 and A as centers (*III* and *IX* in fig. 7). The abnormal ridge produces in addition a narrowing of either the pulmonary or the aortic tract.

Type III—Complete Transposition (fig. 9 *f*). —In this anomaly the aorta emerges from the right ventricle and the pulmonary artery from the left. In most cases the ventricular septum is closed. In a lesser number there is a defect in the anterior portion of the ventricular septum. The arterial trunks ascend from the heart almost parallel to each other. The ostiums of the coronary arteries are usually more transposed, being situated in the posterior sinuses of Valsalva. The coronary arteries are still more abnormal. The thickness of the right ventricle exceeds that of the left. The remainder of the heart is usually normal.

In our interpretation the abnormality here is complete absence of ridge 3 B with no replacement. Ridge 1 A has been destroyed by the abnormal currents created in such a bulbus. The bulbus has then been absorbed without torsions (*V* and *XI* in fig. 7). At the end stages of absorption of such a bulbus currents are established which lead to the production of a proximal bulbar septum, thus completing the ventricular series.

Type IV—Miscellaneous Group.—In this group are included Spitzer's group IV, truncus communis (fig. 9 *g*), truncus solitarius aorticus, truncus solitarius pulmonalis with transposition and transposition with atresia of the tricuspid valve (fig. 9 *h* and *i*). Spitzer's group IV consists of those cases in which a hypoplastic aorta emerges from a small humplike pouch, separated from a large ventricular space into which open both auriculoventricular ostiums, or a common auriculoventricular ostium, and from which emerges the pulmonary artery.

In truncus communis there is not only an abnormality in the absorption of the bulbus as indicated in types I and II but an abnormality in the septum aorticopulmonale trunci (Lev and Saphir [14]). Truncus solitarius aorticus and truncus solitarius pulmonalis are exaggerated forms of type II transposition (Eisenmenger or Fallot types). In transposition with atresia of the tricuspid valve one is dealing with an absence of ridge 3 B, but ridge 1 A is not obliterated. The absorption of a bulbus with such a ridge proceeds as follows: At the distal bulbar ostium less torsion occurs, while at the proximal ostium there is 45 to 90 degrees of back torsion. Because of the absence of ridge 3 B, torsion occurs about ridge 1 A as a center (*IV* and *X*). In addition there is an abnormality in the auriculoventricular ostiums. Normally, besides the bulbar movements occurring in the second phase, there is a shift of the auricular canal to the right (fig. 4). Thus, whereas both auriculoventricular ostiums are originally to the left side of the bulbus at the end of the first phase, the tricuspid orifice passes to the right of the mesocardial portion of the bulbus after the normal movements of the second phase. With this goes an expansion of the tricuspid orifice and of the dorsal wall of the meta-ampule (distal portion of the primitive ventricle). If, therefore, there is an abnormality in the absorption of the bulbus, especially in the type in which rotation occurs about ridge 1 A as a center, then in some cases the tricuspid orifice and the right auricle do not reach their normal definitive position to the right of the bulbus. This results in atresia of the tricuspid valve.

The abnormality in Spitzer's type IV is similar to that just described without the changes in the auricular canal.

13. Saphir, O., and Lev, M.: Am. Heart J. **21**:31, 1941.

14. Lev, M., and Saphir, O.: J. Pediat. **20**:74, 1942.

CRITERIA FOR THE PATHOLOGIC DIAGNOSIS OF TRANSPOSITION

The criteria for the presence of transposition are (1) an abnormal position of the arterial trunks, (2) an abnormal position of the ostiums of the coronary arteries (counterclockwise displacements if one is looking from the aorta into the ventricle), (3) an abnormal topography of the muscle bundles of the right ventricle and (4) an abnormal distribution of the coronary arteries.

The distribution of the coronary arteries has been thoroughly studied by Spitzer.[6] The ramus descendens anterior is taken over by the artery arising from the right sinus of Valsalva, or two branches, one from the right and one from the left, may be present. The posterior descending branch is usually taken over by the artery arising from the left sinus of Valsalva. In some cases the two sinuses may have completely changed branches.

The topography of the muscle bundles of the right ventricle in the normal heart and that in the heart with transposition have been studied by many, and their work is reviewed elsewhere.[1] In our studies we noted two types of topography prevalent in normal hearts (fig. 8). In most cases two distinct muscle bundles are noted. From the septal cusp of the pulmonic valve a muscle bundle obliquely descends over the septum toward the apex. This we have called the septal muscle bundle or band. Near the apex it gives off the moderator band. On the anterior wall of the right ventricle the parietal muscle bundle or band ascends toward the base of the heart in close proximity to the anterior leaflet of the tricuspid valve. The superior portion of the muscle terminates at the septal pulmonic cusp. Its main intermediary portion fuses with the musculature of the septum beneath the septal band. A distinct raphe is noted where the parietal band dips behind the septal band. The inferior portion terminates at the base of the anterior leaflet of the tricuspid valve. In a smaller number of cases there is a mass of musculature forming an arch over the base of the ventricle at the base of the septal cusp of the pulmonic valve, with fibers radiating over the anterior wall of the right ventricle, adjacent to the tricuspid valve and down over the right wall of the septum. This formation apparently represents a fusion of the septal and parietal bands. From the evidence of Keith[4] and Pernkopf and Wirtinger,[7] the formation of the parietal band is related to the formation of the proximal bulbar septum and correlated with the growth of ridge 3 B in mammals. It is thus derived from cushion B, the bulboauricular spur and the counter ridge BO of the ventricular septum. The septal band, from the evidence of Spitzer,[6] Fuchs,[15] Beninghoff,[15] Tandler[16] and Keith,[4] is derived from cushion A (or is an extension of bulbar ridge 1 A).

In transposition of the arterial trunks the septal band is usually hypertrophied and may even form a pseudoseptum (Spitzer). The parietal muscle bundle is usually diminished in size or may be completely absent.

CONCLUSION

Transposition of the arterial trunks may be considered to be produced by an abnormality in the absorption of the bulbus, in the second phase of the development of the heart. This we believe is due to an abnormality in the phylogenetically recently developed bulbar ridge 3 B.

15. Citied by Pernkopf and Wirtinger.[7]

16. Tandler, J., in Keibel, F., and Mall, F. P.: Manual of Human Embryology, Philadelphia, J. B. Lippincott Company, 1912, vol. 2.

Case Reports

PARAPHYSIAL CYST OF THE THIRD VENTRICLE

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The paraphysis is a phylogenetically ancient organ which in former years interested mainly zoologists and embryologists. It was first described by Selenka,[1] in 1890, who found it in embryos of various vertebrates. The gland is best developed in Amphibia. In adults of this class it occurs as a well developed organ. Its location is the anterior portion of the roof of the third ventricle. The paraphysis of Necturus, a member of the class Amphibia, was studied in great detail by Warren,[2] who described it as a lobulated organ composed of anastomosing tubules. The function of this organ is apparently not known. However, it may be mentioned that some authors[3] have accepted a certain homology between the paraphysis of vertebrates and the acoustic organ of ascidians, better known under the popular name "sea squirts". The ascidians (tunicates) represent an interesting group of nonvertebrates which during the larval state of their existence display certain features peculiar to vertebrates, such as a chorda dorsalis and a tubular nervous system. In vertebrates above Amphibia the paraphysis becomes gradually less conspicuous. Rudiments of this organ in the human embryo were first described by Francotte,[4] in 1894. As early as 1909 it was suggested by Sjövall[5] that remnants of the paraphysis may give rise to cysts. In recent years such cysts of presumably paraphysial origin have been described in increasing number, and up to date almost 70 cases have been reported in the literature. With but few exceptions,[6] most of the cases reported in this country appeared in publications pertaining to neurology,[7] although the general pathologist is apt to encounter such cysts in routine autopsy material. An additional case is reported here.

REPORT OF A CASE

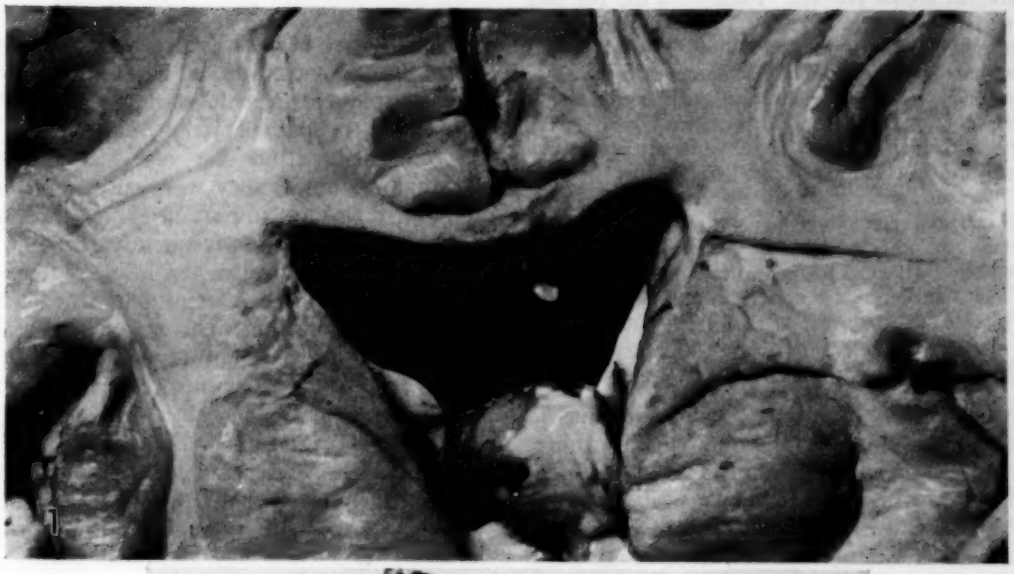
The patient was a woman 29 years old. She was seen late one evening by Dr. H. G. Storner, Olean, N. Y., at her home. She complained of excruciating headaches associated with spells of vomiting. These headaches had begun late the same afternoon. Her pain was severe enough to necessitate two subcutaneous injections of a solution of a morphine salt during the night. The physician made a tentative diagnosis of intracranial neoplasm. The patient died the following morning. Information obtained from relatives disclosed that she had suffered from attacks of headaches for a long time and that they had increased in frequency and severity following extraction of a tooth two and a half months prior to her death.

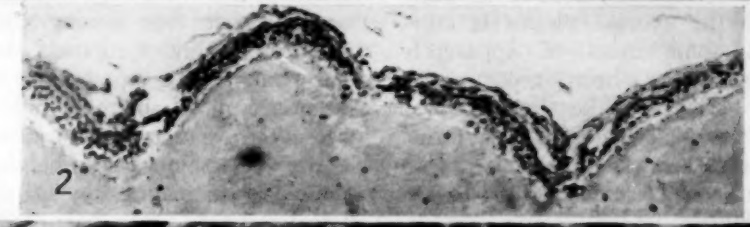
Postmortem Examination (three hours after death).—The organs of the thoracic and abdominal cavities did not disclose any pertinent abnormalities. The brain revealed an approximately normal amount of subarachnoidal fluid. The pia-arachnoid was not thickened. The gyri appeared to be slightly flattened. Coronal sections (fig. 1) of the brain showed marked dilatation of both lateral ventricles. The degree of dilatation was about equal on both sides. In the vicinity of the interventricular foramens of Monro, the roof of the third ventricle presented a cystic mass which extended downward into the cavity of the third ventricle. This cyst measured 1.5 cm. in diameter and contained grayish white viscid fluid. The septum pellucidum above it was extremely thin. The cyst leaned against the foramens of Monro, obviously obstructing the latter. The anterior portion of the third ventricle was enlarged by the cyst, which was suspended from its roof. The aqueductus cerebri and the fourth ventricle were not dilated.

Microscopic Examination of the Cyst.—In addition to the main cyst there were several smaller cystic areas as seen in a multiloculated cyst. The linings of all these cystic areas were about the same (figs. 2 and 3). They consisted of one or two layers of cuboidal cells, some of which were quite flat. A number of the lining cells were distinctly ciliated. The nuclei were fairly large in comparison with the size of the cells and were usually at the base of the cells, away from the ciliated surface. The cytoplasm frequently showed various degrees of vacuolation. Where the vacuolation was extreme, cilia could not be discerned. Indeed, some of these cells had no sharp cell boundaries toward the free surface and appeared to merge with the contents of the cyst. The latter stained moderately acidophilic. Several small concrements which stained bright red with eosin were present in the cyst contents. They were surrounded by polymorphonuclear leukocytes and a few desquamated epithelial cells. Underneath the lining epithelial cells

From the Department of Pathology, Olean General Hospital.

1. Selenka, E.: Biol. Centralbl. **10**:323, 1890-1891.
2. Warren, J.: J. Comp. Neurol. **28**:75, 1917.
3. Krabbe, H. K.: Brain **59**:483, 1936. Selenka.[1]
4. Francotte, P.: Bull. Acad. roy. d. sc. de Belgique **27**:84, 1894.
5. Sjövall, E.: Beitr. z. path. Anat. u. z. allg. Path. **47**:248, 1909.
6. (*a*) Rehbock, D. J.: Arch. Path. **21**:524, 1936. (*b*) Weinberger, L. M., and Boshes, B.: Surgery **13**: 368, 1943. (*c*) Hassin, G. B., and Anderson, J. B.: U. S. Vet. M. Bull. **6**:56, 1930.
7. (*a*) Dandy, W. E.: Benign Tumors of the Third Ventricle of the Brain, Springfield, Ill., Charles C Thomas, Publisher, 1933. (*b*) Zimmerman, H. M., and German, W. J.: Arch. Neurol. & Psychiat. **30**:309, 1933. (*c*) Rinder, C. O., and Cannon, P. R.: ibid. **30**: 880, 1933. (*d*) Stookey, B.: Bull. Neurol. Inst. New York **3**:446, 1934. (*e*) Davidoff, L. M., and Dyke, C.: ibid. **4**:221, 1935. (*f*) McLean, A. J.: Arch. Neurol. & Psychiat. **36**:485, 1936. (*g*) Gardner, W. J., and Turner, O.: ibid. **38**:1055, 1937. (*h*) Shaver, M. R.: ibid. **43**:510, 1940. (*i*) Zeitlin, H., and Lichtenstein, B.: J. Nerv. & Ment. Dis. **91**:704, 1940. (*j*) Larson, C. P.: ibid. **91**:557, 1940.





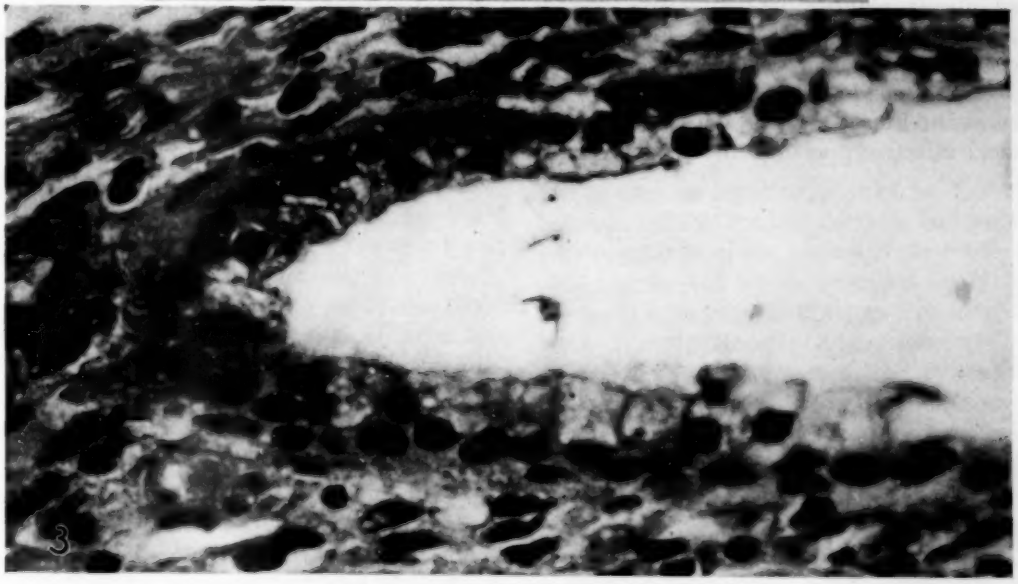


Fig. 1.—Coronal section of the brain showing a cystic mass filling the dilated anterior portion of the third ventricle. Note the marked dilatation of the lateral ventricles.

Fig. 2.—Cyst wall under low power magnification.

Fig. 3.—High power field (dry) showing the lining of one of the additional smaller cysts with partly ciliated epithelium and large macrophages in the adjacent stroma.

there was a stroma which consisted in part of collagenous tissue, varying in thickness, and in part of densely packed, fairly large cells containing clumps of light grayish brown pigment. These cells did not take a sudan stain or a stain for iron. They apparently were macrophages. In one area there was a small tubule lined by cuboidal epithelium. Such tubules, as well as the previously mentioned additional small cystic areas, were thought by Zeitlin and Lichtenstein [7l] to be proof of origin in the paraphysial anlage. Most of the external surface of the cyst had lost its epithelial lining. In some areas, however, one could still discern a single layer of low cuboidal epithelium such as that seen in the ependyma. In other sections the choroid plexus was observed to be attached to the outer surface of the cyst wall. The adjacent tissue of the brain (columnae fornicis) showed a few minute areas of fresh hemorrhage.

COMMENT

The clinical importance of such a cyst lies in its strategic position near the foramens of Monro. Acute impaction in the foramens has been mentioned as a cause of sudden death and was probably the cause of death in the case now described. At the same time the close relationship of such a cyst to the choroid plexus is noteworthy. Indeed, some cysts of apparently paraphysical origin have been reported as choroidal cysts. However the present case illustrates again, as emphasized by other authors, that the cyst while expanding downward into the third ventricle becomes only enfolded by the choroid plexus without actually being part of, or having derived from, the same. Furthermore, Zeitlin and Lichtenstein pointed out that similar cysts containing colloid material are not found in the lateral and fourth ventricles, a fact which can be upheld effectively against the possible origin of such a cyst of the third ventricle from the choroid plexus.

Cysts of this nature—also referred to as neuroepithelial cysts or colloid cysts of the third ventricle—have been described mainly in persons from 20 to 50 years old. They have been three times as frequent in males as in females. About half of all the patients whose cases have been reported died within one year after the onset of symptoms unless they were successfully operated on. The most important symptom is severe paroxysmal headache, frequently altered by a change of the position of the head. It has been suggested that these intermittent headaches are due to temporary encroachment on the foramens of Monro by a more or less freely suspended cyst. Such headaches are frequently accompanied by nausea as well as by papilledema. Stookey [7d] expressed the belief that the headaches are produced by compression of both veins of Galen at their origin with coincident interference with the venous outflow of the choroid plexus of the lateral and third ventricles. Stookey [7d] mentioned further that among patients examined more thoroughly symptoms referable to the diencephalon were found fairly frequently, such as hypersomnia, diabetes insipidus, hyperthermia, adiposity, vasomotor disturbances and others. Ventriculography has been helpful in establishing a diagnosis, as reported by Dandy [7a] and Davidoff and Dyke.[7e] Once diagnosed, these cysts are amenable to surgical intervention. Successful removal of such cysts with cure of the patients has been reported by a number of authors. Weinberger and Boshes [6b] reviewed the literature in 1943.

General Reviews

ARTERIOSCLEROSIS

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THE ANOXEMIA THEORY

(Concluded from page 131)

Excessive Intake of Cholesterol.—The role of alimentary hypercholesteremia in the production of atheromatosis and atherosclerosis is not entirely well established. While the gradual development of persistent and considerable hypercholesteremia in herbivorous animals, especially rabbits, having normally a low level of blood cholesterol, is regularly seen when these animals are put on an abnormal diet containing appreciable amounts of cholesterol, the hematic reactions observed in this respect in carnivorous and omniverous animals placed on a similar diet are distinctly different. Bürger and Winterseel, as well as Bürger and Habs, observed in a human subject given 5 Gm. of cholesterol in olive oil a transitory hypercholesteremia reaching its peak within two hours after the oral introduction of the cholesterol. Similar observations of a transient increase of the blood cholesterol in man after the ingestion of a food rich in fats and cholesterol have been reported (Muller; Barreda; Gardner; Garb; Snapper and Parisel). Corcoran and Rabinowitch stated, on the other hand, that Eskimos living on a meat diet and having normally a high metabolic rate have normal blood lipid values. Page cited Tolstoi as having found that white men on a similar diet over long periods exhibited little change in the level of the blood cholesterol. In proper evaluation of these observations it must be noted that the Eskimos consume relatively little fat (Thomas), and thus there is no valid reason why alimentary hypercholesteremia should develop.

Okey and Stewart, on the other hand, observed in women after administration of a food containing large amounts of cholesterol (egg yolk, butter) a transitory increase in blood cholesterol. Similar observations in man fed powdered egg yolk were recorded by Steiner and Domanski. The cholesterogenic nature of this reaction was demonstrated by the observation that the feeding of soybean lecithin to man caused hypocholesteremia. Flock, Corwin and Bollman obtained sustained hyperlipemia of dietary origin in dogs. Steiner and Domanski also noted such an effect in dogs given a cholesterol-rich food, but the elevation of the blood cholesterol remained within moderate limits and atheromatous reactions were absent. Li, Hough, Monahan and Freeman demonstrated in dogs fed a protein-deficient, high fat–cholesterol diet an increase of the serum cholesterol of 172 to 225 per cent. This reaction was much less marked when fat and cholesterol were given with a diet containing adequate amounts of protein. Reports of persistent alimentary hypercholesteremia in dogs and rats fed a cholesterol-rich diet were published by Corwin and Sperry and Stoyanoff. Whether or not the mildly and persistently increased blood cholesterol not infrequently noted in obese children and adults is of dietary or of endocrine origin has as yet not been determined (Ricketts). The available evidence indicates that the prolonged and excessive intake of cholesterol can elicit persistent, though usually mild to moderate, hypercholesteremia in omnivorous and carnivorous animals.

The earliest manifestation of an interrelation between lipoidal intimal deposits and an alimentary imbalance of the plasma cholesterol is probably represented by the so-called lipoid spots of the aorta appearing as early as two months after birth (Ernst; Görög). These spots and streaks apparently are responses to the relative hypercholesteremia elicited by a milk diet in an organism with an incompletely developed cholesterol metabolism. This conclusion is derived from the fact that the serum cholesterol level of the newborn is low (about 60 mg. per hundred cubic centimeters) (György; Hueck; Muhlbock and Kaufmann; Sperry), which contrasts sharply with the hypercholesteremia prevailing in the maternal blood during pregnancy, when the placenta apparently serves as a filtration membrane and a storage organ for cholesterol (Versé). The blood cholesterol of the infant approximately doubles during the first four months of life (Kube and Ssowlowjew; Knauer) and then rises grad-

ually until it reaches the normal serum cholesterol level of 130 to 230 mg. per hundred cubic centimeters. Thus, according to the figures given before, the human infant behaves in regard to the alimentary intake of cholesterol, which is not inconsiderable, much as does the herbivorous rabbit, in which the serum cholesterol level remains at a low level (50 to 100 mg. per hundred cubic centimeters) throughout life. Studies of Ssowlowjew on suckling rabbits have shown the presence of similar lipoidal deposits in the aortic intima and media, while these were absent in young rabbits which had been on a vegetarian diet for a period of one to two months.

The lipoid spots and streaks observed in the human aorta and the large elastic arteries increase in frequency and extent during the first two decades and tend to disappear thereafter (Lubarsch; Saltykow; Mönckeberg; Schmidtmann; Scheel; Zinserling; Kube and Ssowlowjew). They are present in all children above the age of 5, according to Schmidtmann. Similar changes develop during the same period in the ventricular side of the large mitral leaflet (Martius). The spots and streaks appear as white or white-yellow elongated formations, resembling drops running down a candle, and are not elevated or only slightly elevated above the intimal surface. They first develop just above the aortic cusps, later around the orifices of arteries branching from the aortic arch, in the scar of the ductus arteriosus, on the posterior wall of the aorta, either around or at the lower edges of the orifices of the intercostal and lumbar arteries or between the orifices of the intercostal arteries at the posterior aspect of the aortic wall, at orifices of the large abdominal arteries and, finally, in the lower abdominal portion of the aorta, affecting also the lateral and anterior aspects (Zinserling; Ranke; Lange; Froboese). The lipoidal material appears first in the endothelial cells and extracellularly in the subendothelial layer, but usually also here within histiocytes, especially when little ground substance is present (Zinserling). Fibroblastic proliferation of a mild degree often accompanies these changes (Lubarsch; Schmidtmann). The lipoid spots do not show any progressive degeneration and apparently are reversible (Thorel; Krisch). The cellular proliferation in the periphery of the lipoidal deposit may regress with the ultimate disappearance of the lipoidal material, sometimes leaving a minor focal hyaline thickening of the intima (Zinserling). During midde age both lipoid spots and atherosclerotic lesions may coexist and may even be superimposed on one another (unless such combinations are to be considered different stages of the same process).

A lively controversy still exists concerning some aspects of the causation and the significance of these lesions, especially in regard to whether they are precursors of the atherosclerotic lesions developing in later decades. Beitzke asserted that the lipoid spots are the result of defects in the intima caused by a transitory excessive dilatation of the aortic wall which elicits ruptures in the delicate and unyielding intima. Others (Askanazy; Sanders; Zinserling; Simnitzky) related these lesions to toxic injuries sustained during infectious diseases. Still others (Saltykow; Ranke; Lubarsch; Benda; Anitschkow; Mönckeberg; Kohlhaas; Oberndorfer; Busch; Schmidtmann; Zinserling) regard the lipoid spots as early stages of atherosclerotic lesions, with which they share the same topographic distribution. Ranke claimed that gradual transitions between the lipoid spots and typical atheromas can be demonstrated. Aschoff distinguished sharply between the atherosis of infants and juvenile persons and the sclerotic atherosis of adults. Others (Ribbert; Hueck; Lange; Cramer; Askanazy; Froboese; Westenhöfer; Jores; Beitzke) considered lipoid spots as distinct from atherosclerosis for a number of reasons, such as the difference in the age of occurrence, the irreversibility of atherosclerotic lesions, the presence of productive and degenerative processes in atherosclerotic lesions, the greater distribution of the atherosclerotic changes in the arterial tree and the relative rarity of typical atherosclerosis in children (Benda; Seitz; Chiari; Filatoff and Rachmaninoff).

In the light of the evidence available on the genesis of lipoidal intimal deposits it appears that the differences between lipoid spots and typical atheromas are not fundamental but mainly those of degree and duration. The causative agent and mechanism acting in the production of these manifestations during childhood act evidently with minor intensity and over a limited length of time, while those operative during adult life act through the course of decades and with greater strength.

The available evidence connecting an excessive alimentary intake of cholesterol-containing foodstuffs with the development of atherosclerosis in adults is mainly of circumstantial character (Beneke). Rosenthal, who compiled the data from twenty-eight papers dealing with the relation between diet and the incidence of atherosclerosis in various countries and under different climates, found that a diet high in fat content was almost invariably accompanied by a high

rate of atherosclerosis among its consumers, while diets with a low fat content, such as those consumed by Eskimos and Japanese, were usually associated with a low incidence of atherosclerosis. Investigations of Rubner (in Japanese) and Raab on this subject led to similar conclusions. Grotel, Bykhovskaya, Pavlova, Pokhodilova and Shor found that the degree of atherosclerosis increased parallel with the quality of nutrition, being most pronounced in persons with excessive nutrition, particularly of cholesterol. Rosenthal stated that the intake of large amounts of neutral fat paves the way for the absorption of cholesterol through the intestinal wall. Cramer recorded an unusual incidence of intimal lipoidosis in tuberculous patients kept on a high fat diet. Additional support for the occurrence of alimentary atherosclerosis in man is supplied by the observations of Kuczynski among the inhabitants of the Kirghiz steppe. Kuczynski found that the nomads inhabiting the plain, who daily consume enormous amounts of mare's milk (20 liters) and meat (10 to 20 pounds [4.5 to 9 Kg.]), showed in addition to obesity signs of precocious cholesterosis (extensive atherosclerosis, arcus senilis of the cornea), while the inhabitants of the towns, who consume a more moderate and mixed diet, exhibited a sclerosing hyperplastic type of vascular disease without regressive intimal changes.

Many attempts have been made to prove or to disprove the existence of relations between the cholesterol content of the blood and the development of atherosclerosis by comparing the blood cholesterol content with the condition of the arteries. The results obtained were contradictory, as might be expected from the fact that atherosclerosis is in general a process which takes years to develop and which may persist for many more years after the cessation of the action of the causal agent. The determination of the blood cholesterol content, on the other hand, merely renders information about the momentary condition of the blood, which may be entirely unrelated to the status of the blood prevailing when the vascular lesions were produced. Liebig thus found hypercholesteremia in 77 per cent of his patients with atherosclerosis and essential hypertension and concluded from this observation that cholesterol contributed to the development of the vascular lesions. Steiner and Domanski recorded for patients with coronary arteriosclerosis a serum cholesterol level which fluctuated between 308 and 499 mg. per hundred cubic centimeters, average 355 mg., against values of 214 to 334 mg. per hundred cubic centimeters, average 255 mg., in a normal control group. Davis, Stern and Lesnick reported similar observations in patients with angina pectoris and arteriosclerosis, many of whom showed an increase of all lipid fractions.

Numerous attempts have been made to reproduce atherosclerosis in carnivorous (dog, cat) and omnivorous animals (rat, mouse, chicken) by feeding a diet to which considerable amounts of cholesterol were added. Carnivorous animals normally have retention hypercholesteremia only during pregnancy and after castration (Löwenthal; Neumann and Herrmann; Roemer; Berberich; DaBella; Mancke). Omnivorous and carnivorous animals, like man, absorb and excrete rapidly ingested lipoid material and therefore react with a rapid and transient rise of blood cholesterol (Yuasa). If lipoid is fed over long periods, there is a decrease in the degree of visible lipemia as the organism becomes trained in handling lipoids (Löwenthal).. A combined protein-lipoid diet increases the digestive transitory lipemia in these animals (Bang). While attempts to produce persistent hypercholesteremia in dogs by feeding cholesterol or lecithin in the diet, have been successful (Corwin; Grigaut and L'Huillier; Anitschkow), all efforts to elicit atheromatosis in these animals have so far failed (Wacker and Hueck; Hueck; Kawafura; Adler; Anitschkow). Similar failures were obtained with cats (Cook; Yuasa; Hueck; Cirio; Anitschkow).

The experiments with rats fed a cholesterol-rich diet gave variable results. Numerous observers (Chalatow; Mosebach; Anitschkow and Chalatow; Cook and McCullagh; Chanutin and Ludewig) reported a negative outcome of their experiments, while Saxton and Yuasa recorded occasional or extensive lipoid deposits in the aortic intima. The reason for these divergent results in rats is not apparent from the published data. In feeding mice with various kinds of fats Domagk obtained in addition to amyloid and leukemoid changes in the liver and the spleen fatty degenerations and calcifications of the walls of arteries. Rabl recorded similar results as to arterial lipoidosis and calcinosis in mice fed a cholesterol-containing diet adjusted by the addition of phosphoric acid, ammonium chloride, potassium sulfate, calcium phosphate and sodium acetate to an alkaline or an acid reaction. While Löwenthal was unsuccessful in eliciting atheromatous reactions in normal mice fed a cholesterol-containing normal diet, he succeeded in this respect in castrated mice and in mice fed a protein-cholesterol diet. Corwin mentioned briefly that attempts to produce alimentary atheromatosis in foxes were unsuccessful.

Experimental cholesterol atheromatosis affecting the aorta and the brachiocephalic, coronary, renal, celiac, iliac and femoral arteries was relatively readily and consistently obtained in young chickens by several workers (Uchiyama; Wolkoff; Dauber and Katz). Chickens behave like mammals in regard to their cholesterol metabolism, according to Sperry and Stoyanoff. The cholesterol content of the serum was markedly elevated in chickens fed cholesterol (430 to 2,148 mg. per hundred cubic centimeters) (Dauber and Katz). The ratio of free cholesterol to esters remained within normal limits. The resulting intimal lesion consisted of extracellular and intracellular lipoidal deposits associated with fibroblastic and elastic fibrillar proliferations. Calcifications and ossifications within the foci were not rare. The intimal cushions sometimes encroached markedly on the lumens of large and small arteries, occasionally causing almost complete obliteration. In such severe lesions intimal foam cell proliferations invaded the media, which was often calcified. In addition to lipoidotic changes in various parenchymatous organs, the testes were atrophic, similar to those seen in cholesterol-fed pigeons with atheromatous arterial changes (Kawamura; Yamaguchi). The vascular lesions showed a topographic distribution similar to that of the juvenile lipoid spots seen in man. Dauber and Katz suggested that the difference in the localization of the atheromatous reactions in man and chickens is probably attributable to the fact that different functional and anatomic strains are exerted on various parts of the aorta of these two species. These investigators noted that the pulmonary arteries and veins were free from atheromatous lesions, while the vasa vasorum of the aorta were filled with foam cells. This observation led Dauber and Katz to believe that the migratory foam cells clogging the vasa vasorum initiate the degenerative changes (necrosis, deposition of cholesterol, fibrosis, calcification) in the vascular walls.

For the proper evaluation of these observations it is necessary to point out that Sperry and Stoyanoff found markedly increased serum cholesterol in some of their chickens kept on a normal diet, while Kraus noted that chickens start to show intimal lipoidal deposits in the large arteries from their second year of life. These lipoidal foci are located mainly in the ascending and abdominal portions of the aorta and involve both the intima and the media. Kraus concluded from this evidence that reports of experimental production of atheromatosis in chickens are of uncertain interpretation.

Hesse and Wolkoff, who reported the occurrence of severe and generalized atheromatosis and calcinosis of the arterial tree in 3 parrots kept in captivity for many years and fed a diet of nuts and eggs, considered these manifestations as results of alimentary cholesterosis. It may be mentioned, however, that Cook failed to observe such changes in a parrot receiving a diet fortified with cholesterol and that similar arterial changes are commonly seen in old parrots (Nieberle; Pallaske; Wolkoff; Fox).

Among the herbivorous animals, monkeys, goats, guinea pigs and rabbits have been used for the experimental production of alimentary cholesterol atheromatosis. All attempts to elicit such lesions in monkeys failed (Duff; Corwin). Chalatow fed 3 goats egg yolks for two and a half to five months and obtained a threefold increase of the serum cholesterol in a pregnant female goat and in a young male goat, while the blood cholesterol level of an old male goat remained unchanged. The aorta of the young male goat showed many intimal intracellular lipoidal deposits located especially around the orifices of the aortic branches. The aortas of the older animals were without any significant changes. Chalatow fed guinea pigs egg yolk for a relatively short time without obtaining atheromatous arterial reactions. Anitschkow, who repeated these experiments and extended them for up to one hundred and eighty-three days, produced typical atheromatous intimal responses in the aorta. Pseudoxanthoma cells located between the endothelium and the internal elastic membrane had taken up lipoid material primarily deposited in the interstices of the intima. Similar results in guinea pigs were recorded by Bailey, Cook and Schönheimer. Cook and McCullagh, who fed guinea pigs cholesterol in arachis oil for up to two hundred and eleven days and obtained marked hypercholesteremia, found, on the other hand, only minor intimal lipoid deposits in the aorta and the large vessels.

The great majority of experiments on alimentary cholesterol atheromatosis were performed on rabbits. In the original experiment of Ignatowski, which started the experimentation on alimentary cholesterol atheromatosis, rabbits received a diet of milk and egg yolk, which is rich in cholesterol. After the demonstration of cholesterol as the active principle in this diet subsequent investigators followed several lines of dietary management in their experimental studies on rabbits. One group of workers (von Leersum; Warischtew; Anderson; Newburgh and Squier; Newburgh and Clarkson; Knack;

Meeker and Kesten; Stuckey; Fahr; Duff; Starokadomsky; Saltykow; Steinbiss; Clarkson and Newburgh) continued to feed a diet containing cholesterol as a part of a proteinic base, such as egg yolk, milk, brain, powdered liver and meat. The majority of workers used cholesterol dissolved in an oily medium (sunflower oil, olive oil, linseed oil, arachis oil, cottonseed oil, sodium oleate), which was either mixed with the basal diet or given by stomach tube (Versé; Katz; Sanders, Megibow and Carlen; Leary; Liebig; Anitschkow; Aylward and Stott; Rosenthal; Bollman and Flock; Matsunami; Friedberg and Hurwitz; Duff; Schmidtmann; Schönheimer; Hirsch and Weinhouse; Weinhouse and Hirsch; Wolkoff; Zinserling). A few investigators (Diez; Shapiro; Chuma; Nisi) fed cholesterol in the forms of hydrous wool fat, whereas only the exceptional student added dry cholesterol to the ordinary diet (Wacker and Hueck; Scarff; Knack). Atherosclerosis was successfully produced by all these dietary procedures, but relatively least readily by the last-mentioned method.

The alleged production of intimal lipoidal deposits following the administration of lecithin (Remesow; Seemann) has to be charged to the cholesterol usually present in preparations of lecithin of animalic origin (Schönheimer). Lecithin prepared from soybeans apparently has no atherosclerotogenic properties. Wesselkin's results with lecithin were negative. The feeding of vegetable protein (soybeans, gluten flour) (Freyberg; Stuckey) or of defatted liver (Kon) or of various kinds of oils and fats free from cholesterol (Versé; Duff) was equally ineffective. Diez, on the other hand, asserted that the feeding of spermaceti, which does not contain any cholesterol, caused hypercholesteremia. Raab stated that whenever steps have been taken to remove the cholesterol from the proteinic matter the feeding of this material fails to elicit atheromatous lesions.

It must be mentioned in this connection that the feeding of a protein diet free or almost free from cholesterol to rabbits has repeatedly resulted in vascular changes of a sclerotic-calcinotic or sclerotic-lipoidotic type. Steinbiss reported that in rabbits kept on a diet of powdered liver or testis there developed, simultaneously with osteoporosis, only degenerative changes in the muscular and elastic elements of the aortic media, associated with fibrosis and calcinosis. Similar lesions were sometimes seen in the small arteries of the kidney, the liver and the spleen. Nuzum, Osborne and Sansum repeated the experiments of Steinbiss and observed in their rabbits the development of hypertension and arteriosclerotic lesions consisting of swelling and calcification of the intimal ground substance and hyalinization and calcification of the elastic fibrils and of the muscle cells of the media. Nuzum, Seegal, Garland and Osborne observed during subsequent experiments performed on rabbits which received for two years a pure oat diet and in which hypertension developed transitory swelling of the intimal ground substance followed by deposition of cholesterol and hyalinization and fibrosis of the intima. Through cyclic repetition of these processes there occurred a layered thickening of the intima with lipoidal-hyaline deposits. In both the liver-fed and the oat-fed rabbits acidosis developed as a result of the diet, and Nuzum and his co-workers attributed to it the production of the hypertensive and atherosclerotic reactions. They considered a dietary cholesterol intake as unessential for the production of atheromatous lesions, on the basis of these observations. Meeker and Kesten recently took up again the feeding of rabbits with a cholesterol-free protein (defatted casein), added to the regular diet, and observed in the rabbits thus treated the development of mild to moderate hypercholesteremia and of arteriosclerotic changes in the aorta and the coronary vessels. When the casein was replaced in the diet by soybeans, the production of atherosclerotic lesions was prevented by the iodine normally contained in soybeans.

In this connection reference may be made to experiments made by Newburgh and Clarkson and by Anderson. These experimenters fed rabbits a diet 50 per cent of which was powdered lean beef meat. The rabbits obtained thereby a total daily cholesterol intake of 28 mg., and typical atheromatous lesions developed in the aorta in addition to intimal and medial calcifications. Newburgh and Clarkson were inclined to disregard the possible role which the small amount of ingested cholesterol may have played in the production of the vascular responses and expressed the belief that these reactions were the result of the protein diet. These observations suggest the possibility that in rabbits certain types of protein diet may elicit under proper experimental conditions of unknown nature hypertensive, hypercholesteremic and atheromatous or atherosclerotic or arteriosclerotic-calcinotic reactions ensuing from disturbances of the calcium and cholesterol metabolism.

These observations are of special interest as certain workers (Schmidtmann and Hüttich; Schmidtmann; Westphal; Deicke; Fahr; Matusita; von Leersum, and Schönheimer) have asserted that the feeding of cholesterol, egg yolk,

hydrous wool fat or liver results in the development of hypertension, which is causally connected with the hypercholesteremia produced, since renal changes are absent. Schmidtmann proposed that the hypertension is attributable to a sensitizing effect exerted by the cholesterol on epinephrine. The hypertensive reactions paralleled the atheromatous vascular changes, according to Schmidtmann. These observations, however, were not confirmed by others (Tregubow; Shapiro and Seecof; Thölldte; Shapiro, Shepard and Seecof; Katz, Sanders, Megibow and Carlen). The cardiac hypertrophy noted in cholesterolized rabbits by Katz, Sanders, Megibow and Carlen and by Friedberg and Hurwitz could not be traced to systemic hypertension but was attributed to the myocardial ischemia caused by the atheromatosis of the coronary arteries, which, on the other hand, did not elicit abnormal electrocardiographic reactions (Nyboer, Bruger and Rabson). These experimental data indicate in connection with the clinical experience that hypercholesteremia and hypertension are not causally interrelated.

The increase in the serum cholesterol content of the cholesterolized rabbits may be considerable (up to twenty times the normal amount) and is not accompanied by any consistent changes in the ratio of free to esterified cholesterol. While this reaction is accompanied by elevation of the levels of the other lipoids and lipids of the serum (Weinhouse and Hirsch; Wacker and Hueck; Bollman and Flock; Page and Bernhard), the cholesterol increase is disproportionally higher than that of any other of the fat substances. The rise in serum cholesterol during the first two months under this dietary management is a gradual and progressive one. After two to three months the movement levels off and is followed by a drop in spite of continued oral administration of cholesterol. The serum cholesterol level finally becomes stationary at a point several times the normal value. It is noteworthy that this drop cannot be prevented by increasing the dose of cholesterol ingested but can be slowed by increasing the amount of oil given (Versé; Weinhouse and Hirsch; Reineck; Kirchgessner; Rohrschneider). There seems to develop first a balance between absorption, on one hand, and excretion and storage, on the other. After all possible depots for lipoid storage in the organism are filled to capacity, there follows apparently a metabolic adaptation of the body to the increased intake of cholesterol, resulting in either more rapid excretion of the cholesterol or decreased absorption of it from the intestine.

It is noteworthy that the type of oil used as a solvent influences the speed of absorption of cholesterol and of the development of hypercholesteremia. Linseed oil seems to be most effective in this respect (Versé).

The deposition of cholesterol in the arterial walls is usually preceded by considerable storage of lipoids in various organs (liver, adrenal glands, kidneys, spleen, lungs) and in the reticuloendothelial system when the usual method of feeding large daily doses of cholesterol is used. The chronic effects of this thesaurosis elicit cirrhotic changes in the liver (Versé; Broun and Muether). Lipoid deposits in the corneas (arcus senilis) of cholesterolized rabbits are frequent (von Poppen; Rohrschneider; Versé; Kolen; Chuma; Schönheimer; Nakanonim; Nakanonim and Koboschi; Kawamura; Wada, and others). These corneal changes recede partly after cessation of the oral ingestion of cholesterol. Marked differences exist in the degrees to which the various lipoids and lipids are stored in the different organs (Aylward and Stott). The organic retention of glycerides appears to be independent of that of cholesterol and to be influenced by the choline, protein and oil contents of the diet.

Experiments of Anitschkow, of Chuma and of Zinserling and Krinitzky have demonstrated that the development of organic lipoidosis and of high hypercholesteremia is not prerequisite and essential to the development of alimentary atherosclerosis in rabbits. When Anitschkow fed rabbits with small amounts of diluted milk and egg yolks and when Chuma and Zinserling and Krinitzky administered small quantities of hydrous wool fat to rabbits over periods of two years, they noted isolated and mild atheromatous reactions in the aorta in the absence of storage phenomena in the parenchymatous organs and in the presence of only minor hypercholesteremia. The causative and hematic conditions present in these experiments resemble closely those observed in many instances of human atherosclerosis. Anitschkow showed, moreover, that a short and intensive course of oral treatment with cholesterol is sufficient to elicit atheromatous lesions, which are formed in the presence of minor organic lipoidosis and which persist and are newly formed for several months after the arrest of treatment and when the serum cholesterol level has returned to normal. The absence of hypercholesteremia is thus not proof that such a disturbance did not furnish the causal background of atherosclerosis.

Atheromatous deposits in the arteries appear first and are most marked in the ascending por-

tion of the aorta and then around the orifices of the intercostal arteries. Later they extend to the abdominal portion and to the larger arteries —the carotid, subclavian, iliac, renal, splenic, intestinal, renal and coronary arteries (Duff; Anitschkow; Hurwitz and Friedberg; Wolkoff; Schmidtmann; Scarff; Katz, Sanders, Megibow and Carlen; Versé; Cook and McCullagh; Leary). The large and small branches of the pulmonary artery are frequently involved; while with severe nodular xanthomatous atheromatosis the veins also show lesions. Schönheimer observed endothelial and subendothelial deposition of lipoid in the intima of the inferior vena cava and in the portal vein, which was similar to that seen occasionally in man with high hypercholesteremia (Benda).

The atheromatous formations start with the appearance of lipoidal matter in the subendothelial ground substance and in foam cells of this region or with foamy swelling of the endothelial cells. The subendothelial foam cells are in part derivatives of the proliferated xanthomatous endothelial cells, but mainly they evidently originate from histiocytic cells, especially in those cases in which they appear beneath an apparently intact endothelial lining. Anitschkow and also Leary, however, insist that the foam cells are exclusively of histiocytic or reticuloendothelial origin. The internal elastic membrane may show fraying and proliferation and in severe lesions is penetrated by foam cells invading the media. In advanced changes there may be foam cell transformation of the endothelial lining of the vasa vasorum as well as accumulations of foam cells in the spaces around these vessels. Medial calcifications beneath the foam cell intimal cushion are occasionally seen (Danisch; Hueper).

Early regressive changes in these lesions were observed by Stuckey and by Krylow during the first six months after the cessation of cholesterol feeding. Versé reported fibromuscular transformation of the plaques with central calcification in a rabbit which survived the dietary treatment by four hundred and twenty-five days. Wada recorded similar reactions in rabbits after hydrous wool fat had been fed. Anitschkow, who studied the aortas of rabbits one hundred and one to eight hundred and fifteen days after the discontinuation of the cholesterol management, found the first signs of regressive changes in the atheromatous lesions in the ascending and upper thoracic portions of the aorta; such reactions did not appear in the atheromas of the abdominal portion of the aorta until much later. During the regressive changes the lipoid first disappears from the foam cells and remains only in the periphery of the plaque, while the center is replaced by fibrous tissue. Collagenous and elastic fibrils develop between the foam cells. The fat released from the disintegrating foam cells separates into large coalescing globules of neutral fat and cholesterol crystals. The neutral fat is resorbed, while the cholesterol crystals remain. Calcium granules then appear in the deeper parts of the plaques, and the fibrosis increases (Anitschkow; Duff). New formation of capillaries is only rarely seen in the atheromas (Anitschkow). Saltykow observed similar regressive changes in rabbits maintained continuously on a milk diet. The development of aneurysms of the aorta in the region of the arch on the basis of these regressive intimal and medial changes was reported by Liebig, by Wesselkin and by Leary and Weiss. In the case reported by Leary and Weiss a dissecting aneurysm was found similar to those frequently observed in the rabbit's aorta after the injection of epinephrine hydrochloride (Fischer; Erb; Külbs; Ziegler, Kaiserling; Schirokogoroff).

Numerous experiments have been performed in attempts either to accentuate or to accelerate the development of dietary cholesterol atheromatosis in rabbits or to impair or to suppress this process. Ssolowjew succeeded in aggravating the local deposition of cholesterol in exteriorized parts of carotid arteries, which were placed in skin sleeves. These arteries were traumatized when the rabbits threw their heads violently backward during daily attempts to introduce a stomach tube. These abrupt movements of the neck produced tears in the elastic membranes. The defects thus produced were filled in by lipoid-containing foam cells. While Schmidtmann recorded aggravation of the vascular lesions in cholesterolized rabbits through administration of vitamin D, Harrison observed that vitamin D sclerosis in rabbits rendered the affected parts relatively immobile and that this immobility seemed to make the sclerosed foci less susceptible to the subsequent development of atheromatous lesions. Meeker and Kesten reported that a diet high in soybean flour diminished the incidence and the degree of experimental atheromatosis, while a diet rich in defatted casein augmented them. The attempts of Jobling and Meeker to accelerate or to increase the development of cholesterol lesions in the aortas of rabbits failed when the following procedures were employed: Intravenous injection of streptococcus toxin, feeding of ammonium hydroxide, production of artificial fever, intravenous injection of peptone, production of anaphylactic shock and intravenous injection of uric acid. These steps were taken

so as to elicit one of two apparently primary changes in the intima important in the development of experimental and human atherosclerosis: (1) a change in the permeability of the wall of the vessel in the neighborhood of the internal elastic membrane which prevents the normal passage of large molecules, such as proteins and lipids, from the intima into the media without interfering with the passage of water and crystalloids; (2) an increase in the permeability of the intima without other injury to the vessel wall. In both instances it was thought that there may occur a deposition of relatively large amounts of lipids and of coagulated and finally hyalinized protein. Similar experiments of Thiersch, who tried to enhance atheromatosis by producing bacterioallergies, likewise failed.

Shapiro noted that thyroidectomized, splenectomized or gonadectomized rabbits exhibited increased susceptibility to cholesterol atheromatosis. This susceptibility was especially marked in thyroidectomized animals. Moehlig's attempts to attain this goal in cholesterolized rabbits by repeated injections of a solution of the posterior lobe of the pituitary gland were successful. Injections of the solution alone proved to be ineffective. A similar aggravating effect was exerted on the atheromatosis by the thyrotropic factor of the pituitary gland (Bruger and Fitz; Sperry).

Various means have been employed in the many attempts made to prevent or to cure the cholesterol atheromatosis of rabbits. Alcohol, recommended by Leary as an antidote against cholesterol atheromatosis in man, was tried by Eberhard in cholesterolized rabbits. It was found that if the cholesterol was dissolved in 20 per cent alcohol, the absorption of the cholesterol from the intestine was accelerated and the rise of the blood cholesterol level was more rapid and higher than when cholesterol was added dry to the basal diet. While there were no gross differences in the appearance of the atheromatous aortas of the two groups, the lesions in the alcohol series exhibited smaller intimal cushions which extended less often into the media. The significance and the finality of these observations, however, were regarded by Eberhard as doubtful. Schaffir fed egg yolk and alcohol to rabbits and observed increased aortic atheromatosis and medial calcifications. Thiersch reported similar results in rabbits equally treated with cholesterol and alcohol. Chaiika noted that lactic acid and oleic acid when fed to rats along with cholesterol increased the resulting hypercholesteremia but prevented deposition of fat in the aorta. These acids also intensified the uptake of lipids by the cells of the reticuloendothelial system. After Bürgi had contended that chlorophyll exerted a favorable effect on arteriosclerosis and hypertension, Blumer, Gordonoff and Reznikoff and Gordonoff fed rabbits cholesterol in oil and a preparation of chlorophyll and observed only minor fibrotic thickenings in the aortic intima and absence of lipemia and lipoidosis of the Kupffer cells. Malisoff and Hueper claimed that the administration of potassium thiocyanate reduced the severity and the incidence of atherosclerosis in thyroidectomized rabbits fed cholesterol. The mechanism of this protective action is, according to Malisoff, not clear, as it is uncertain whether the thiocyanate heightens the dispersion of the cholesterol colloids in the blood and the tissues or changes the permeability of the vascular wall (Westphal), as thiocyanates are colloidochemical antagonists of cholesterol (Guttmann).

Attempts of Hesse to influence favorably the cholesterol atheromatosis of rabbits by the administration of an ethyl ester of ricinoleic acid and of its silicic acid adduct were unsuccessful. Equally unsuccessful were the efforts of Flexner, Bruger and Wright to prevent the development of hypercholesteremia and atheromatosis in cholesterolized rabbits by the injection of massive doses of ascorbic acid and thiamine singly or combined, as well as those of Thiersch in connection with the administration of dehydrocholic acid.

While the experimental results of Kesten and Silbowitz suggested that the administration of soybean lecithin or of choline in an amount equivalent to that in lecithin mitigated the development of cholesterol atheromatosis in rabbits, the studies of Steiner, of Himsworth and of Baumann and Rusch showed that the administration of even large doses of choline had no influence on the cholesteremia and atheromatosis of cholesterolized rabbits. Steiner reported, however, as a suggestive finding that choline might aid in the mobilization and resorption of lipoids from atheromatous lesions produced in the aorta. Although Huber, Broun and Casey reported that lipocaic prevented the development of hypercholesteremia and atheromatosis in cholesterolized rabbits, subsequent investigations (Vermeulen, Allen, Clark, Julian and Dragstedt; Vermeulen, Dragstedt, Clark, Julian and Allen; Dragstedt, von Prohaska, Clark and Julian) did not confirm this claim. Wright contended, on the other hand, that the administration of a deproteinized pancreatic extract to cholesterolized rabbits retarded the development of atheromatosis and caused a striking transitory lowering of the blood cholesterol level. Ludden, Bruger and Wright showed that estradiol dipropionate and testosterone propionate had no in-

fluence on the cholesterol content of the blood and the aorta when given over a period of one hundred days. However, when female rabbits fed cholesterol were treated with either of these substances they showed inhibition of the hypercholesteremia associated with almost complete absence of cholesterol deposits in the aorta. No such results were seen in male rabbits identically treated. Gonadectomy did not alter the response of normal female rabbits, but the protective action of these substances toward cholesterosis was abolished after castration. These observations are important, as atherosclerosis and medial calcinosis are in general less frequent in women than in men. The frequency of coronary arteriosclerosis is thus four and nine-tenths times as great in men as in women (Levy and Boas). Similar percental conditions exist in regard to thromboangiitis obliterans, suggesting a protective action related to estrogen.

The majority of experimental investigators in this field have used either iodides, which in many respects resemble cyanides in their biologic and physicochemical action (Westphal), or thyroid preparations. Iodides have been employed for many years in the treatment of arteriosclerosis and hypertension, mainly on an empiric basis. Numerous explanations have been advanced for the alleged favorable results obtained clinically in patients. Guggenheimer and Fisher stated that iodine has a vesodilative effect (Sée and Lapicque; Krause; Bogolepoff; Rose; Huchard; Eloy; Bloom; Kochmann; Liebe). This assertion was contradicted (Boehm and Berg; Barkan and Prikk; Orth; Eppinger and Hess; Freund and König). Some have contended that iodides decrease the volume of erythrocytes and thus lower the blood viscosity, thereby relieving hypertension (Romberg; Müller and Inada; Deusch and Frowein). This claim also was disputed (Determann and Bröking; Alwall; Buchholtz; Boruttau). Adam even found an increase in blood viscosity after iodide medication. Others attributed the beneficial effect of iodide on atherosclerosis to an increase in pulse volume and thereby an improvement in circulation (Lehndorff) or to complex endocrine and metabolic reactions (Hildebrandt; Hesse; Wegelin; Wada; Mansfeld; Barkan) or to a direct action on the atheromatous material (Ungar; Loeb) or to an effect on vegetative nervous centers (Schittenhelm and Eisler; Abelin; Sturm and Veil) or to an inhibition of the accumulation of cholesterol deposits in the arterial walls (Damrau). Schottmüller, as well as Masing, on the other hand, did not see any favorable effect from iodides in arteriosclerosis, while Turner and Steiner observed either no effect on the serum cholesterol level of patients with various diseases or exceptionally even an increase after prolonged introduction of iodides.

These contradictory observations in man, which are in part evidently due to the fact that arteriosclerotic patients were subjected to medication with iodine without regard to etiology or type, contrast sharply with those made on rabbits treated for dietary cholesterol atheromatosis either with an inorganic iodide (potassium iodide or sodium iodide) or with various organic iodides (Liebig; Meeker, Kesten and Jobling; Bruger and Fitz; Murata and Kataoka; Seel and Creuzberg; Friedland; Page and Bernhard; Turner and Khayat; Strauss; Turner and Bidwell; Masson; Page; Turner; Ungar; Thiersch). All investigators found that iodides given together with cholesterol prevent or mitigate the development of cholesterol atheromatosis. Organic iodides, especially compounds of fatty acids, were apparently more effective than inorganic compounds because they remain in the blood longer than the inorganic iodides (Bröking). Their effect is apparently not dependent on a reduction in the serum cholesterol level, although this reaction is commonly seen after the administration of iodides. Page and Bernhard, who used a diiodide of ricinsterolic acid, found in their iodide-treated series higher lipemia than that present in the controls which received cholesterol only. While fatty acid–iodine compounds retain their effectiveness in thyroidectomized rabbits (Ungar), the effect of inorganic iodides depends on the presence of the thyroid gland, for their protective action is abolished after the removal of this gland (Turner and Khayat). The complexity of these reactions is indicated by the fact that the favorable effect of iodides is not a simple metabolic oxidation effect exerted through the thyroid gland or a hypocholesteremic effect but is probably connected with a stabilizing action on the equilibrium of the plasmatic colloids. Inasmuch as iodide medication has no influence on lesions once established, this type of therapy is of purely preventive and not therapeutic character (Thiersch).

A similar protective action in rabbits is demonstrable when thyroid preparations are given together with cholesterol (Zon; Murata and Kataoka; Friedland; von Baló; Menne, Beeman and Labby). In cholesterolized rabbits there was a reduction or an absence of hypercholesteremia as well as an absence or a reduction of atheromatous manifestations. In contrast to prolonged iodide medication, which finally failed to check the development of a hypercholesteremia and sometimes even accentuated this reaction, the administration of thyroid preparations maintained the hypocholesteremic effect unchanged

(Turner and Steiner). The inconstancy of the iodides in this respect may be attributable to the fact that prolonged iodine medication may elicit a hypothyroidotic, myxedematous state following a primary hyperfunctional response.

Leary and Raab recommended as a practical measure for the control of atherosclerosis in man a restriction of the dietary intake of cholesterol through a lessened consumption of butter, egg yolks and meat. This stand appears to be well taken so as far as it concerns persons whose consumption of these substances is highly excessive and persons with disturbances of the fat metabolism (diabetes mellitus, obesity, essential xanthomatosis, psoriasis).

Many investigators maintained that the experimental alimentary cholesterol atheromatosis of rabbits differs in several fundamental aspects from the human atherosclerosis and that therefore no reliable conclusions can be drawn from the observations made on rabbits as to the causative mechanism and the etiologic factors of this condition in man (Klotz; Duff; Hirsch and Weinhouse). It is often maintained that the cholesterol atheromatosis of rabbits resembles the lipoid spots in man or the xanthomatous vascular lesions seen in essential xanthomatosis but not typical atherosclerosis. It is pointed out that the atheromatosis of rabbits occurs only in association with and as a sequela of generalized lipoidosis, that it is obtained only when rabbits are fed an abnormal diet, that it differs in the topographic distribution of its lesions from the human variety, that the histologic structure of the atheroma varies from that of the human type, that the same measures fail to produce similar lesions in carnivorous animals, that in rabbits the cerebral arteries are spared, while they are often affected in man, and that the atheromas of rabbits do not ulcerate, with development of thrombotic deposits.

Although it must be conceded that the lipoid metabolism of the rabbit differs in certain respects from that of man, this does not detract from the fact that in both species disturbances of the cholesterol content of the plasma are associated with atheromatous arterial lesions of essentially the same type. Hypercholesteremia and instability of the plasmatic colloidal lipoidal solution represent the common denominator for the development of the organic lipoidoses and the vascular atheromatosis seen in the acute alimentary form of cholesterosis of the rabbit and the accelerated types of metabolic cholesterol disturbances accompanying essential xanthomatosis, diabetes mellitus and hypothroidism. The hematic and vascular reactions seen in rabbits fed small amounts of cholesterol over long periods are not unlike those observed in many instances of the ordinary type of human atherosclerosis. It is relatively immaterial as far as the immediate causative mechanism in man is concerned whether or not an excessive alimentary intake of cholesterol plays a significant role in the production of the atherosclerotic lesions, as there are many other causes for the imbalance in the plasmatic colloidal lipoidal solution. The evidence available in this respect in man, together with the fact that alimentary factors are apparently involved in the development of atherosclerosis in other omnivorous animals, makes such a connection not unlikely. It can scarcely be maintained that at the present stage of knowledge of human nutrition any exact statements can be made as to how normal or abnormal the diet of natural and processed foodstuffs of the average human being is, especially if consideration is given to the differences in constitution and environment, and as to what long range effect it exerts on the plasmatic lipoidal equilibrium and the status of the vascular system.

The differences in the topographic distribution of the atheromatous lesions in man and rabbits are evidently in part due to differences in localizing static factors. This conclusion is supported by recent observations made by Wilens, who was able to produce a shift of the atheromatous manifestations from the ascending part of the aorta into the abdominal part by keeping cholesterolized rabbits for five hours daily in an erect position, imitating thereby the static conditions present in man. It is likely, moreover, that differences in the course and in the structure of the various arteries of man and rabbits exert a definite influence in this respect, as is apparent from observations made in man and rabbits as well as in other species. Moschcowitz laid main emphasis on the local fixation and the external resistance of the vascular wall, which control the expansive mobility of the vessel, as the cause of the localization of atherosclerotic lesions under the influence of increased intravascular hydrostatic pressure. In support of his thesis he pointed to the following observations: 1. The earliest patches of aortic sclerosis are at or near the origin of intercostal vessels which fix the posterior wall of the aorta. 2. The abdominal part of the aorta, which is fixed against the spine, is more involved than the thoracic. 3. Dural vessels are most markedly involved when lying in bone. 4. The anterior aspect of the aorta is less involved than the posterior, as the latter is fixed against the spine. 6. Radial arteries show the most marked sclerosis where they lie directly against the radius. 7. Aortic patches appear ear-

liest in places corresponding to upper and lower borders of bodies of vertebrae. 8. Pulmonary arteries lying against rigid bronchial cartilage are most involved. 9. The left coronary artery, embedded in firmer muscle, is more affected than the right one.

Aschoff and many others asserted that the areas around the orifices of intercostal arteries and at arterial bifurcations are exposed to increased mechanical strain and thus are subject to degenerative changes which favor the infiltration of plasma and the precipitation of cholesterol. Harrison maintained that the movements of a vessel determine the localization of cholesterol lesions in animal as well as probably also in human arterial disease, since local immobility appears to render the area in which it is present less susceptible to subsequent atheromatous lesions. Oberndorfer found in man no atheromatosis in arteries overlying joints (popliteal space), though these parts of vessels are often in motion.

Ranke pointed out that lumbar lordosis developing during life as a result of erect posture influences the blood current in the aorta and may explain the higher incidence of atherosclerosis in the abdominal part of the aorta. Atherosclerosis, according to this investigator, involves earliest and most extensively those vascular regions in which the compensation to the hematic push–tension of the wall is disturbed or lowered for some reason (as in the concave parts of curves, at bifurcations) and where the range of compensation is lowered. The concave part of the aortic arch is much more stretched by the dislocation of the heart when a full stomach is present than the convex side. Distention in width means a shortening pull in length. These influences interfere with the nutritive current in the wall by impairing the outflow of tissue lymph, and thus edema develops which causes tension of the fibrils, which in turn impairs the attachment of the intima to the media. At such spots atherosclerosis develops. The predominating location of atheromatosis in the abdominal part of the aorta is explained by the fact that only here a sufficient longitudinal shift of the aorta occurs causing diffuse changes, while the nodular lesions are the result of abdominal pressure exerted on the abdominal portion of the aorta by the abdominal wall. The location of atheromas on the posterior aspect of the aorta is attributable to the circumstance that the aorta is not extendable in this region, as it is fixed to the spine (Ranke).

Albrecht noted that the internal carotid artery embedded in the petrous bone shows more fatty degeneration and calcification than the common carotid artery or the axillary artery. Smetana attributed the early occurrence of atheromatosis in the aortic bulb to the action of a functional strain surpassing that of other parts of the vessel, while the distribution of the vasa vasorum does not play a role in this respect.

Differences in the structure of the arterial walls of man and rabbits, particularly in the thickness of the intima and in the amount and the distribution of the elastic tissue, probably exert an additional modifying influence on the distribution of the atheromatous lesions. The importance of this factor is evident from the following statements. Duff proposed that cholesterol is precipitated at sites where the vascular wall is altered, i. e., where primary focal destruction of the muscle fibers or edematous swelling of the subendothelial ground substance occurs. On the basis of observations made during experiments in which trypan blue was injected into rabbits, he contended that the permeability of the intima of the aorta of the rabbit to trypan blue is uniform throughout and that the local accumulations of the dye in the aortic wall are related to the distribution of the vasa vasorum from which the dye leaks out into the media. The intensification of the dye around the orifices of the intercostal vessels is attributed to increased tissue lymph flow through these regions rather than to slowing of it. The same causative mechanism, it is held, accounts for the accumulation and precipitation of cholesterol in these areas.

Rosenthal, on the other hand, favored the concept that slowing of the tissue lymph stream in the intima with the elastic barrier acting as a hindrance to permeation of the media by lipoids is responsible for the accumulation of the lipoids in the intimal tissues, but he did not provide a sufficient reason for their precipitation. A similar conception was proposed by Anitschkow, who stated that the occurrence of cholesterol deposits depends on general conditions (status of the blood and the nutritive fluid permeating the vascular walls) and on local conditions determining the exact site of the deposits (local injury of the wall caused by change in the ground substance). The proliferative responses accompanying this process he considered as secondary. Pekelharing noted that the rhythmic changes in the intravascular pressure with the pulse exert the smallest pressure where the vascular wall yields most. The local loss of elasticity thus furnishes the focus for intimal proliferation following local vascular dilatation. A similar concept is propounded by Dormanns and Emminger, who claimed that the marked appearance of atherosclerosis in the ascending and thoracic parts of the aorta in the presence of syphilitic

aortitis is attributable to the formation of local aortic dilatations due to the destruction of elastic tissue.

Leary stated that the first lesions occur at sites of special mechanical stress which causes, however, no injury to the intima. With the closing of the aortic cusps the ascending part of the aorta is subjected momentarily to greater stress than any other part of the arterial system. As the proximal portions of the coronary arteries share in this stress, this mechanism accounts for the selective frequency of sclerosis in these parts of the coronary arteries. A static aspect of atherosclerosis and mediocalcinosis of the arteries in the lower extremities receives some support from the observations of Man and Peters and of Keys and Butt, who found that on standing there is an increase in the lipoid and protein contents of the blood of these vessels as watery components leak through the vascular walls under the influence of increased static pressure causing increased permeability of the vascular walls. Beneke pointed out that the blood flow is normally not uniform, but whirl formation occurs especially at curves, orifices of branches and bifurcations. The increased stress entailed thereby provides the cause for the localization of atheromas.

While the factual observations concerning the distribution of atheromatous lesions at various special points of the arterial tree are correct, none of the numerous reasons offered in explanation of the findings appears to be satisfactory from a colloidochemical standpoint, and many of the arguments advanced disregard entirely the colloidal aspect of the production of the lesions. If slow blood flow and stagnation of tissue fluid represent an important feature of the accumulation and precipitation of lipoids in the vascular wall, it must be expected that not the arteries but the veins would be the location of atheromatosis. In fact, atheromatosis is found most marked in those vessels in which a fast blood flow prevails. If the distribution and the plugging of the vasa vasorum with lipoid cells are of significance, again the venous walls should be the most frequent site of atheromatosis, as the veins possess a well developed network of vasa vasorum which extends into the intima and have a slow blood flow favoring the arrest of lipophages on the vascular walls and their invasion of the vasa vasorum. The actual distribution of the atheromatous reactions is fundamentally different.

If the preferred sites of atheromatous deposits in the arterial tree are analyzed from a hemodynamic point of view, it becomes apparent that they represent spots where the uniform blood flow is acutely disturbed, i. e., where turbulence of the blood occurs. It is obvious that at the base of the aorta is an area in which the blood is exposed to frequent and violent vibration due to the recoil of the blood during the period of diastole as well as during the systolic impact of the cardiac blood on the aortic blood column. The resulting whirl formation, doubtless, is accentuated whenever the aortic wall is studded with small retractions which break the marginal blood current and which are formed as the result of syphilitic scarring of the media. The blood in the proximal parts of the coronary arteries is subjected to a similar mechanism when during systole the intramyocardial branches are obliterated by the contracting myocardial tissue and the coronary circulation comes practically to a temporary standstill. The blood is then pressed into the extramyocardial parts of the coronary vessels and especially into their proximal portions, setting up there violent whirl formations. Any local dilatation of the vascular wall, whether caused anatomically by abnormal focal changes in the wall or resulting functionally from a one-sided circumscribed fixation or external resistance, exerts a similar effect on the blood current. Natural or pathologic narrows of vessels, such as passages through bone or the presence of orifices of branches, bifurcations and curves, also increase the turbulency of the blood stream in circumscribed parts of the arterial tree.

Hellwig recently pointed out that atheromatosis of the mitral valve is apparently due to the fact that the colloidally dispersed lipoids are precipitated on the ventricular side of the mitral leaflet because the stability of the plasmatic cholesterol solution is disturbed by the vigorous systolic percussion of the mitral leaflets, with precipitation of the lipoids. The phenomenon is comparable as to mechanism to the lipoid flocculation elicited by shaking in the Kahn test. Similar changes were reported by Hueper in connection with his experiments with polyvinyl alcohol, which is precipitated out of solution on shaking. It seems reasonable to assume that such vibratory disturbances of the plasma are responsible for the distribution of the atheromatous lesions in the vascular tree at the aforementioned sites.

The deposition of the precipitated lipoids depends in part on such local factors and in part on static conditions, as the precipitated material tends to settle in the dependent portions, i. e., in the abdominal part of the aorta of the full

grown person. This influence will be much less pronounced or even absent in the young person in whom such static conditions are either nonexistent or only mildly active. The experiments of Wilens on cholesterolized rabbits kept in an erect position support the soundness of this reasoning.

It is not unlikely that the different physicochemical conditions of the arterial and venous blood, particularly the differences in hydrogen ion concentration, may play an important part in controlling the stability of the plasmatic colloidal equilibrium and in accounting for the fact that atheromatosis is in general restricted to the arterial part of the vascular tree. The relations existing in this respect may be analogous to those influencing the precipitation of calcium from the plasma, as here also the alkaline state of the arterial blood favors such a process in the arterial wall and in the left side of the heart, while the more acid state of the venous blood tends to keep calcium salts in solution. Inasmuch as necrotic tissue usually has an alkaline reaction and in view of the fact that cholesterol membranes in the interface of the plasma and the intima or within the intima favor the development of degenerative changes in the vascular wall, it appears to be possible that disturbances of the cholesterol metabolism and of the plasmatic colloidal lipoidal equilibrium play an important causal role in the production of the mediocalcinosis of the peripheral arteries, particularly those of the lower extremities.

The importance of a primary colloidal status of the atheromatogenic substances in the plasma for the production of the vascular atheromatous lesions is illustrated additionally by the fact that these agents when present in the plasma in noncolloidal dispersion, i. e., as a coarse emulsion, do not elicit foam cell intimal reactions but foreign body giant cell granulomas which especially affect the small vessels and capillaries of the lung but occasionally also other parts of the vascular tree, such as the testicular venus plexus (Hirsch and Weinhouse; Hirsch; Seemann; Merkulow; Hueper). The lung apparently acts here as a filtration organ in which the globules of lipoids or macromolecular carbohydrates are arrested and reacted on.

COLLOID PLASMATIC DISTURBANCES: B. LIPOIDAL TYPE

Adam, H.: Ztschr. f. klin. Med. **68**:177, 1909.

Addis, T., and Oliver, J.: The Renal Lesions in Bright's Disease, New York, Paul B. Hoeber, Inc., 1931.

Alpers, B. J.: Arch. Neurol. & Psychiat. **42**:1173, 1939.

Alwall, N.: Acta med. Scandinav. **101**:83, 1939.

Anderson, H.: Arch. Int. Med. **37**:297, 1926.

Anitschkow, N.: Beitr. z. path. Anat. u. z. allg. Path. **56**:379, 1913; **57**:201, 1914; **59**:306, 1914; **70**:265, 1922; Virchows Arch. f. path. Anat. **220**:233, 1915; **249**:73, 1924; Klin. Wchnschr. **4**:2233, 1925; **13**: 1259, 1934; Verhandl. d. deutsch. path. Gesellsch. **20**:149, 1925; **23**:473, 1928.

——and Chalatow, S.: Zentralbl. f. allg. Path. u. path. Anat. **24**:1, 1913.

Apollonio, C.: Beitr. z. path. Anat. u. z. allg. Path. **3**:290, 1888.

Arning, E., and Lippmann, A.: Ztschr. f. klin. Med. **89**:107, 1920.

Aschoff, L.: Virchows Arch. f. path. Anat. **235**:152, 1921; Beihefte z. Med. Klin. (no. 1) **26**:1, 1930.

Astwood, E. B.: J. Pharmacol. & Exper. Therap. **78**: 79, 1943.

——Sullivan, J.; Bissell, A., and Tyslowitz, R.: Endocrinology **32**:210, 1943.

Aylward, F. X., and Stott, W.: Biochem. J. **31**:2055, 1937.

Bailey, C. H.: Proc. Soc. Exper. Biol. & Med. **13**:60, 1915.

Baker, L. E., and Carrell, A.: J. Exper. Med. **45**:305, 1927.

von Baló, J.: Beitr. z. path. Anat. u. z. allg. Path. **102**:341, 1939.

Balzer, F.: Arch. Physiol. **3-4**:65, 1884.

Bang, I.: Biochem. Ztschr. **91**:86, 104, 111 and 122, 1920.

Barkan, G.: München. med. Wchnschr. **79**:621, 1932.

Barreda, P.: Klin. Wchnschr. **13**:290, 1934.

Baumann, C. A., and Rusch, H. P.: Proc. Soc. Exper. Biol. & Med. **38**:647, 1938.

Baumann, T.: Klin. Wchnschr. **14**:1743, 1935.

Bayer, G., and Gaisböck, F.: Wien. med. Wchnschr. **74**:2001, 1924.

Beitzke, H.: Virchows Arch. f. path. Anat. **267**:625, 1928; **275**:532, 1930.

Bell, E. T.: Am. J. Path. **10**:705, 1934.

Benda, C.: Virchows Arch. f. path. Anat. **254**:600, 1925.

Beneke, R.: Frankfurt. Ztschr. f. Path. **28**:407, 1922; Beitr. z. path. Anat. u. z. allg. Path. **87**:285, 1931; Verhandl. d. deutsch. path. Gesellsch. **35**:251, 1935.

Beutel, A.: Strahlentherapie **69**:400, 1941.

Bianchi, E.: Med. d. lavoro **29**:213, 1938.

Black, H. A., and Kampmeier, R. H.: Colorado Med. **31**:262, 1934.

Blackman, S. S.: Bull. Johns Hopkins Hosp. **55**:1, 1934.

Bloom, D.; Kaufman, S. R., and Stevens, R. A.: Arch. Dermat. & Syph. **45**:1, 1942.

Bloor, W. R.: J. A. M. A. **119**:1018, 1942.

Blotner, H.: New England J. Med. **203**:709, 1930.

Blum, F.: Schweiz. med. Wchnschr. **71**:1612, 1941.

Blumer, C.; Gordonoff, T., and Reznikoff, L.: Arch. f. exper. Path. u. Pharmakol. **173**:42, 1933.

Boehm, R., and Berg, F.: Arch. f. exper. Path. u. Pharmakol. **5**:329, 1876.

Boggs, T. R., and Morris, R. S.: J. Exper. Med. **11**: 553, 1909.

Bollman, J. L., and Flock, E. V.: Arch. Path. **28**:762, 1939; Proc. Am. Soc. Exper. Path., 1941.

Bork, K.: Virchows Arch. f. path. Anat. **262**:646, 1926.

Boruttau: Ztschr. f. exper. Path. u. Therap. **18**:203, 1916.

Bourneville: Arch. d. Neurol. **16**:37, 97, 241 and 346, 1903.

Bröking: Ztschr. f. exper. Path. u. Therap. **8**:125, 1911.

Bronstein, I. P.: J. A. M. A. **100**:1661, 1933.

Bross, K.: Virchows Arch. f. path. Anat. (supp.) **227**: 145, 1920.

Broun, G. O., and Muether, B. O.: J. A. M. A. **118**: 1403, 1942.
Bruger, M.: J. Biol. Chem. **108**:463, 1935.
——and Fitz, F.: Arch. Path. **25**:637, 1938.
——and Rosenkrantz, J. A.: J. Clin. Endocrinol. **2**:176, 1942.
——Wright, I. S., and Wiland, J.: Arch. Path. **36**:612, 1943.
Brunner, W.: Klin. Wchnschr. **14**:1853, 1935.
Buchholtz: Arch. f. exper. Path. u. Pharmakol. **82**:30, 1918.
Bürger, M.: Verhandl. d. Gesellsch. f. Verdauungskr. **14**:88, 1938; Klin. Wchnschr. **17**:1780, 1938; Verhandl. d. deutsch. path. Gesellsch. **31**:88, 1938.
——and Habs, H.: Klin. Wchnschr. **6**:2221, 1927.
——and Schlomka, G.: ibid. **7**:1944, 1928.
Bürgi, E.: München. med. Wchnschr. **74**:2019, 1927.
Buschke, A.; Klopstock, E., and Peiser, B.: Med. Klin. **20**:345, 1924.
Calvin, J. C., and Goldberg, A. H.: Am. J. Dis. Child. **41**:1066, 1931.
Chaiika, T. V.: Arkh. biol. nauk **57**:74, 1940.
Chaikoff, I. L.; Entenman, C.; Changus, G. W., and Reichert, F. L.: Endocrinology **28**:797, 1941.
Chalatow, S. S.: Zentralbl. f. allg. Path. u. path. Anat. **25**:197, 1914; Virchows Arch. f. path. Anat. **207**: 452, 1912; **272**:691, 1929.
Chanutin, A., and Ludewig, S.: J. Biol. Chem. **102**:57, 1933.
Chiari, H.: Verhandl. d. deutsch. path. Gesellsch. **9**:340, 1905.
Cholesterolemia and Thyroid Disorder, editorial, J. A. M. A. **103**:113, 1934; J. Clin. Endocrinol. **3**:526, 1943.
Chuma, M.: Virchows Arch. f. path. Anat. **242**:275, 1923.
Cirio, L.: Virchows Arch. f. path. Anat. **269**:739, 1928.
Clark, H. C.: Am. J. Dis. Child. **59**:353, 1940.
Clarkson, S., and Newburgh, L. H.: J. Exper. Med. **43**:595, 1926.
Collens, W. S., and Wilensky, N. D.: J. A. M. A. **109**:2125, 1937.
Cook, R. P.: Biochem. J. **30**:1630, 1936; Nutrition Abstr. & Rev. **12**:1, 1942.
——and McCullagh, G. P.: Quart. J. Exper. Physiol. **29**:283, 1939.
Corcoran, A. C., and Rabinowitch, I. M.: Biochem. J. **31**:343, 1937.
Corwin, W. C.: Arch. Path. **26**:456, 1938.
Craig, L. S.; Lisser, H., and Soley, M. H.: J. Clin. Endocrinol. **4**:12, 1944.
Damrau, F.: M. Rec. **144**:373, 1936.
Danisch, F.: Klin. Wchnschr. **7**:289 and 337, 1928; Beitr. z. path. Anat. u. z. allg. Path. **79**:333, 1928.
Dauber, D. V., and Katz, L. N.: Arch. Path. **34**:937, 1942; **36**:473, 1943.
Dauksys, J.: J. Missouri M. A. **32**:466, 1935.
Davis, D.; Stern, B., and Lesnick, G.: Ann. Int. Med. **11**:354, 1937.
Davis, H. H., and Warren, S.: Arch. Path. **16**:853, 1933.
De Courcy, J. L.: J. Med. **9**:9, 1928.
Degwitz, R.: Klin. Wchnschr. **9**:2336, 1930.
Deicke, O.: Krankheitsforsch. **3**:399, 1926.
De Langen, C. D.: Acta med. Scandinav. **97**:427, 1938.
Determann: Deutsche med. Wchnschr. **34**:871, 1908.
——and Bröking: ibid. **38**:994, 1912.
Diez, S.: Beitr. z. path. Anat. u. z. allg. Path. **83**:74, 1929.
Dirr, K.: Klin. Wchnschr. **18**:91, 1939.
Domagk, G.: Klin. Wchnschr. **17**:1780, 1938.
——and von Dobeneck, P.: Virchows Arch. f. path. Anat. **290**:385, 1933.
Dominguez, R.: J. Exper. Med. **46**:463, 1927.
Domisch: Klin. Wchnschr. **6**:1781, 1926.
Dormanns, E., and Emminger, E.: Virchows Arch. f. path. Anat. **295**:525, 1935.
Dragstedt, L. R.: J. A. M. A. **114**:29, 1940; Am. J. Physiol. **129**:P348, 1940.
——Clark, D. E.; Julian, O. C.; Vermeulen, C., and Goodpasture, W. C.: Surgery **8**:353, 1940.
——Goodpasture, W. C.; Vermeulen, C., and Clark, D. E.: Am. J. Physiol. **126**:P479, 1939.
Dry, T. J., and Hines, E. A.: Ann. Int. Med. **14**:1893, 1941.
Dublin, L. I., and Lotka, A. J.: Twenty-Five Years of Health Progress, New York, Metropolitan Life Insurance Co., 1937, p. 319.
Duff, L.: Arch. Path. **20**:81 and 259, 1935; **21**:161, 1936.
Duncan, A. G.: J. Ment. Sc. **77**:332, 1931.
Dungal, N. P.: Lancet **1**:1354, 1936.
Dupee, C.; Johnson, V.; Marchello, A.; Wilner, W., and Freeman, L. W.: Federation Proc. **3**:8, 1944.
Eberhard, T. P.: Arch. Path. **21**:616, 1936.
Ehrentheil, O., and Weis-Ostborn, W.: Ztschr. f. Immunitätsforsch. u. exper. Therap. **36**:356, 1923.
von Eiselsberg: Deutsche Ztschr. f. Chir. **73**:47, 1904; Arch. f. klin. Chir. **49**:207, 1895.
Engelberg, H., and Newman, B. A.: J. A. M. A. **122**: 1167, 1943.
English, J. P., and Willius, F. A.: Arch. Int. Med. **71**:594, 1943.
Eppinger, H.: Verhandl. d. deutsch. path. Gesellsch. **31**:51, 1938.
Epstein, A. A.: J. Exper. Med. **20**:334, 1914; J. A. M. A. **69**:444, 1917; Am. J. M. Sc. **154**:638, 1917; **163**:167, 1922; Arch. f. Verdauungskr. **44**:31, 1928.
Epstein, E.: Virchows Arch. f. path. Anat. **281**:152, 1931.
Epstein, E. Z., and Greenspan, E. B.: Arch. Int. Med. **58**:860, 1936.
Ernst, P.: Beitr. z. path. Anat. u. z. allg. Path. **63**: 141, 1916.
Fagge, C. H.: Tr. Path. Soc. London **24**:242, 1873.
Fahr, T.: Verhandl. d. deutsch. path. Gesellsch. **15**:234, 1912; Klin. Wchnschr. **13**:609, 1934.
Falta, W.: Die Erkrankungen der Schilddrüse, in Mohr, L., and Staehelin, R.: Handbuch der inneren Medizin, ed. 2, Berlin, Julius Springer, 1927, vol. 4, pt. 2, p. 1035.
Fenz, E., and Zell, F.: Klin. Wchnschr. **36**:1133, 1936.
Ferraro, A.; Jervis, G. A., and Flicker, D. J.: Arch. Path. **32**:723, 1941.
Filatoff and Rachmaninoff: Jahrb. f. Kinderh. **19-20**: 209, 1883.
Fischer, W.: Deutsches Arch. f. klin. Med. **97**:230, 1909.
Fishberg, A. M.: J. A. M. A. **82**:463, 1924.
Fleischmann, W., and Shumacker, H. B., Jr.: Bull. Johns Hopkins Hosp. **71**:175, 1942.
——Shumacker, H. B., Jr., and Wilkins, L.: Am. J. Physiol. **131**:317, 1940.
——and Wilkins, L.: J. Clin. Endocrinol. **1**:799, 1941.
Flexner, J.; Bruger, M., and Wright, I. S.: Arch. Path. **31**:82, 1941.
Flock, E. V.; Corwin, W. C., and Bollman, J. L.: Am. J. Physiol. **123**:558, 1938.
Fox, C.: Lancet **2**:688, 1879.
Freund, H., and König, W.: Arch. f. exper. Path. u. Pharmakol. **133**:317, 1928.

Freyberg, R. H.: Arch. Int. Med. **59**:660, 1937.
Friedberg, L., and Hurwitz, M.: Federation Proc. **1**:26, 1942.
Friedland, I. B.: Ztschr. f. d. ges. exper. Med. **87**:683, 1933.
Froboese: Zentralbl. f. allg. Path. u. path. Anat. **31**:225, 1920-1921.
Futcher, T. B.: Ann. de dermat. et syph. **7**:887, 1906; Am. J. M. Sc. **130**:939, 1905.
Gallego, J. D.: Arch. f. Gewerbepath. u. Gewerbehyg. **8**:124, 1938.
Garb, J.: Ann. Int. Med. **24**:241, 1943.
Gardner, J. A.: Brit. M. J. **2**:392, 1932.
Gaté, J.; Chanial, G.; Vallet, A., and Humbert, P.: Ann. de dermat. et syph. **9**:465, 1938.
Geiling, E. M. K.; Jensen, H., and Farrar, G. E.: Insulin, in Heffter, A.: Handbuch der experimentellen Pharmakologie, Berlin, Julius Springer, 1935, vol. 5, p. 279.
Ghalioungui, P., and Zell, F.: Arch. f. exper. Path. u. Pharmakol. **185**:71, 1937.
Gibbs, C. B. F.; Buckner, E., and Bloor, W. R.: New England J. Med. **209**:384, 1933.
Gilbert, G. G.: Arch. Int. Med. **68**:591, 1941.
Gildea, E. F.; Man, E. B., and Peters, J. P.: J. Clin. Investigation **18**:739, 1939.
Görög, D.: Virchows Arch. f. path. Anat. **287**:602, 1933.
Goldmann, E.: Beitr. z. klin. Chir. **18**:595, 1897.
Gordy, S. T., and Trumper, M.: J. A. M. A. **110**:1543, 1938; Indust. Med. **9**:231, 1940.
Goreczky, L., and Kovats, J.: Biochem. Ztschr. **314**: 208, 1943.
Greene, A. M.: Arch. Int. Med. **67**:114, 1941.
Grigaut, A., and L'Huillier, A.: Compt. rend. Soc. de biol. **73**:304, 1912.
Gross, P., and Jacox, H. W.: Am. J. M. Sc. **203**:673, 1942.
Grotel, D. M.; Bykhovskaya, E. E.; Pavlova, M. M.; Pokhodilova, M. G., and Shor, V. G.: Klin. Med. **18**:34, 1940.
Gruenfeld, G., and Seelig, M. G.: Arch. Path. **17**:546, 1934.
Grütz, O.: Klin. Wchnschr. **17**:1780, 1938; Verhandl. d. deutsch. path. Gesellsch. **31**:81, 1938; Verhandl. d. Gesellsch. f. Verdauungskr. **14**:81, 1938.
Guggenheimer, H., and Fisher, I. L.: Ztschr. f. d. ges. exper. Med. **54**:114, 1927.
Guttmann, E.: Klin. Wchnschr. **6**:1808, 1927.
Gwynne, C. N.: Rep. Soc. Study Dis. Child. **5**:318, 1905.
György, P.: Klin. Wchnschr. **3**:483, 1924; Zentralbl. f. Kinderh. **112**:283, 1926.
Hamperl, H.: Virchows Arch. f. path. Anat. **271**:147, 1929.
Handovsky, H.: Klin. Wchnschr. **3**:1354, 1924; **10**: 1158, 1931; Arch. f. exper. Path. u. Pharmakol. **134**: 191, 1928; **137**:264, 1928; **159**:383, 1931.
——and von Trossel, I.: Skandinav. Arch. f. Physiol. **49**:145, 1926.
Harbitz, F.: Norsk mag. f. lægevidensk. **86**:321, 1925; **97**:695, 1936; **98**:1317, 1937; **99**:1250, 1938; Arch. Path. **4**:507, 1927.
Harrison, C. V.: J. Path. & Bact. **36**:447, 1933.
Haythorn, S. R.; Taylor, F. A.; Crago, H. W., and Burrier, A. Z.: Am. J. Path. **12**:283, 1936.
Heckscher, H.: Biochem. Ztschr. **158**:417 and 422, 1925.
Heine, J.: Verhandl. d. deutsch. path. Gesellsch. **27**: 78, 1934; Beitr. z. path. Anat. u. z. allg. Path. **94**:412, 1935.
Hellwig, C. A.: Am. Heart J. **24**:41, 1942.
Herman, K.: Orvosi hetil. **78**:1208, 1934.
Herrmann, E., and Neumann, J.: Wien. klin. Wchnschr. **25**:1557, 1912.
Hertzka, E.: Berl. klin. Wchnschr. **18**:567, 1881.
Herzog, E.: Beitr. z. path. Anat. u. z. allg. Path. **85**:707, 1931.
Hess, F. O.: Verhandl. d. deutsch. Gesellsch. f. Kreislaufforsch. **7**:57, 1934; Klin. Wchnschr. **13**:714, 1934.
Hesse, E.: Klin. Wchnschr. **18**:502, 1939.
Hesse, M.: Virchows Arch. f. path. Anat. **261**:225, 1926.
Heyn, F.: Arch. f. Psychiat. **41**:49, 1905.
Himsworth, H. P.: Acta med. Scandinav. (supp.) **90**: 158, 1938.
Hines, E. A., Jr., and Barker, N. W.: Am. J. M. Sc. **200**:717, 1940.
Hirsch, E. F.: Arch. Path. **20**:665, 1935; **21**:764, 1936; **25**:35, 1938.
——and Weinhouse, S.: ibid. **30**:1079, 1940; Physiol. Rev. **23**:185, 1943.
Hoelzer, H.: Beitr. z. path. Anat. u. z. allg. Path. **104**:289, 1940.
Hoffmeyer, J.: Nord. med. **9**:365, 1941.
Hofmeister, E.: Beitr. z. klin. Chir. **11**:441, 1894.
Horn, H., and Finkelstein, L. E.: Am. Heart J. **19**: 655, 1940.
Huber, M. J.; Broun, G. O., and Casey, A. E.: Proc. Soc. Exper. Biol. & Med. **37**:441, 1937.
Hueck, W.: Verhandl. d. deutsch. path. Gesellsch. **20**: 18, 1925; Zentralbl. f. allg. Path. u. path. Anat. **23**:454, 1912.
Hueper, W. C.: Arch. Path. **38**:93 and 350, 1944; **40**:51, 1945.
Hughes, F. W. T., and Perry, C. B.: Bristol Med.-Chir. J. **46**:219, 1929.
Hunt, H. M.: New England J. Med. **201**:659, 1929.
Hurwitz, M., and Friedberg, L.: Arch. Path. **34**:875, 1942.
Hurxthal, L. M.: Arch. Int. Med. **53**:762 and 825, 1934.
——and Hunt, H. M.: Ann. Int. Med. **9**:717, 1935.
——and Simpson, H. N.: J. Clin. Endocrinol. **1**:450, 1941.
Ignatowski, A. I.: Izviest. Imp. Voyenno-Med. Akad. **16**:154, 1908.
Isenschmid: Pathologische Physiologie der Schilddrüse, in Bethe, A.; von Bergmann, G.; Embden, G., and Ellinger, A.: Handbuch der normalen und pathologischen Physiologie, Berlin, Julius Springer, 1930, vol. 16, pt. 1.
Jobling, J. W., and Meeker, D. R.: Arch. Path. **21**: 293, 1936.
Joël, E.: Klin. Wchnschr. **3**:269, 1924.
Johnson, V.; Freeman, L. W.; Longini, J., and Loewy, A.: Federation Proc. **3**:22, 1944.
Joslin, E. P.: Ann. Int. Med. **4**:54, 1930; Trauma and Diabetes Mellitus, in Brahdy, L., and Kahn, S.: Trauma and Disease, ed. 2, Philadelphia, Lea & Febiger, 1941, p. 536; The Treatment of Diabetes Mellitus, ed. 6, ibid., 1937.
Katsch, G., and Krainick, H. G.: Klin. Wchnschr. **18**:436, 1939.
Katz, L. N.; Sanders, A.; Megibow, R. S., and Carlen, S.: Am. J. M. Sc. **200**:731, 1940.
Kauvar, A. J.: J. Clin. Endocrinol. **1**:955, 1941.
Kennedy, T. H.: Nature, London **150**:233, 1942.
Kesten, H. D., and Silbowitz, R.: Proc. Soc. Exper. Biol. & Med. **49**:71, 1942.

Keys, A., and Butt, H. R.: Arch. Int. Med. **63**:165, 1939.
Kimmelstiel, P.: Virchows Arch. f. path. Anat. **282**: 402, 1931.
Kirch, E.: Beitr. z. path. Anat. u. z. allg. Path **70**: 75, 1922.
Kirchgessner, G.: Klin. Wchnschr. **13**:976, 1934.
Klotz, O.: Am. J. Path. **10**:700, 1934.
Knack, A. V.: Virchows Arch. f. path. Anat. **220**:36, 1915.
Knauer, H.: Klin. Wchnschr. **8**:1745, 1929; Ergebnisse der Lipoidstoffwechselforschung mit besonderer Berücksichtigung der Verhältnisse im Kindesalter, Berlin, S. Karger, 1928.
Kofler, L., and Fischer, R.: Arch. f. exper. Path. u. Pharmakol. **141**:105, 1929.
Kolen, A. A.: Virchows Arch. f. path. Anat. **263**:46, 1927; **272**:679, 1929.
Kollert, V., and Rezek, P.: Virchows Arch. f. path. Anat. **262**:838, 1926.
——and Grill, H.: Ztschr. f. d. ges. exper. Med. **49**: 522, 1926.
——Kofler, L., and Susani, O.: ibid. **45**:682, 1925.
Koppenhöfer, G. F.: Virchows Arch. f. path. Anat. **297**:271, 1936.
Kraus, H.: Arch. f. exper. Path. u. Pharmakol. **179**: 537, 1935.
Krisch, H.: Virchows Arch. f. path. Anat. **230**:191, 1921.
Krylow, D.: Compt. rend. Soc. de biol. **79**:397 and 399, 1916.
Kube, N., and Ssolowjew, A.: Frankfurt. Ztschr. f. Path. **40**:302, 1930.
Kuczynski, B.: Klin. Wchnschr. **4**:39, 1925.
Kusnetzowski, N. J.: Zentralbl. f. allg. Path. u. path. Anat. **32**:534, 1921-1922.
Landé, K. E., and Sperry, W. M.: Arch. Path. **22**: 301, 1936.
Lane, G. C., and Goodman, J., Jr.: Arch. Dermat. & Syph. **32**:377, 1935.
Lapowski, B.: Arch. Dermat. & Syph. **11**:701, 1925.
Lasch, F.: Ztschr. f. d. ges. exper. Med. **42**:548, 1924; Klin. Wchnschr. **13**:1534, 1934.
Lauritzen, K.: Virchows Arch. f. path. Anat. **279**:603, 1930.
Leary, T.: New England J. Med. **205**:231, 1931; Arch. Path. **17**:453, 1934; **21**:459, 1936; **29**:665, 1940; **32**:507, 1941; **37**:16, 1944; Am. Heart J. **16**:549, 1938; West. J. Surg. **48**:537, 1940; J. A. M. A. **124**:385, 1944.
von Leersum, M.: Virchows Arch. f. path. Anat. **217**: 452, 1914.
Lehndorff, A.: Arch. f. exper. Path. u. Pharmakol. **76**:224, 1914.
Lehnherr, E. R.: New England J. Med. **208**:1307, 1933.
Lehzen, G., and Knauss, K.: Virchows Arch. f. path. Anat. **116**:84, 1889.
Lerman, J.: J. A. M. A. **117**:349, 1941.
Letterer, E.: Verhandl. d. Gesellsch. f. Verdauungskr. **14**:12, 1938; Verhandl. d. deutsch. path. Gesellsch. **31**:12, 1938.
Levy, H., and Boas, E. P.: J. A. M. A. **107**:97, 1936.
Lewey, F. H.: Ann. Int. Med. **15**:869, 1941; J. Indust. Hyg. **23**:415, 1941.
Li, T.-W.; Hough, V. H.; Monahan, E. P., and Freeman, S.: Federation Proc. **2**:50, 1943.
Lichtenstein, L., and Epstein, E. Z.: Arch. Int. Med. **47**:122, 1931.
Liebe, S.: Arch. f. exper. Path. u. Pharmakol. **171**: 426, 1933.
Liebig, H.: Klin. Wchnschr. **8**:1516, 1929; **10**:475, 1931; **20**:538, 1941; Arch. f. exper. Path. u. Pharmakol. **159**:265 and 359, 1931; **175**:409, 1934.
Lisa, J. R.; Magiday, M.; Galloway, I., and Hart, J. F.: J. A. M. A. **120**:192, 1942.
——Magiday, M., and Hart, J. F.: ibid. **118**:1353, 1942.
Llosa Ricketts, G.: Rev. peruana de pediat. **1**:1, 1942.
Loeb, O.: Arch. f. exper. Path. u. Pharmakol. **69**:108, 1912.
——and Michaud: Biochem. Ztschr. **3**:367, 1907.
Löwenthal, K.: Beitr. z. path. Anat. u. z. allg. Path. **61**:564, 1915-1916; **79**:497, 1928; Verhandl. d. deutsch. path. Gesellsch. **20**:137, 1925; **21**:209, 1926; Frankfurt. Ztschr. f. Path. **34**:145, 1926; Klin. Wchnschr. **5**:478, 1926; Virchows Arch. f. path. Anat. **251**:108, 1926.
Longini, J., and Johnson, V.: Am. J. Physiol. **140**: 349, 1943.
Ludden, J. B.; Bruger, M., and Wright, I. S.: Endocrinology **28**:999, 1941; Arch. Path. **33**:58, 1942.
McArthur, C. S.: Biochem. J. **36**:559, 1942.
McDonald, R.: Arch. Ophth. **20**:839, 1938.
Mackenzie, C. G., and Mackenzie, J. B.: Endocrinology **32**:185, 1943.
Mackenzie, J. B., and Mackenzie, C. G.: Federation Proc. **1**:122, 1942.
——Mackenzie, C. G., and McCollum, E. V.: Science **94**:518, 1941.
Malisoff, W. M.: Proc. Soc. Exper. Biol. & Med. **35**:356, 1936.
Man, E. B., and Peters, J. P.: J. Clin. Investigation **12**:1031, 1933.
Mancke, R.: Arch. f. exper. Path. u. Pharmakol. **149**: 56, 1930.
Marchand, F.: Verhandl. d. Kong. f. inn. Med. **21**:23, 1904.
Maresch, R.: Ztschr. f. Heilk. **19**:249, 1898.
Marine, D.: Arch. Path. **28**:65, 1939.
Martius, K.: Frankfurt. Ztschr. f. Path. **5**:515, 1910; **15**:135, 1914.
Masing, E.: Fortschr. d. Therap. **7**:193, 1931.
Mason, R. L.; Hunt, H. M., and Hurxthal, L.: New England J. Med. **203**:1273, 1930.
Masson, P.: Schweiz. med. Wchnschr. **71**:1042, 1941.
Matusita, T.: Jap. J. M. Sc., IV. Pharmakol. **12**:38, 1940.
Medvei, C. V.: Klin. Wchnschr. **11**:414, 1932.
Meeker, D. R., and Jobling, J. W.: Arch. Path. **18**: 252, 1934.
——and Kesten, H. D.: ibid. **31**:147, 1941; Proc. Soc. Exper. Biol. & Med. **45**:543, 1940.
——Kesten, H. D., and Jobling, J. W.: Arch. Path. **20**:337, 1935.
Member, S.; Ehrlich, S. B., and Bruger, M.: Proc. Soc. Exper. Biol. & Med. **46**:560, 1941.
Menne, F. R.; Beeman, J. A., and Labby, D. H.: Arch. Path. **24**:612, 1937.
Merkel, H.: Beitr. z. path. Anat. u. z. allg. Path. **104**:332, 1940.
Merkulow, G. A.: Virchows Arch. f. path. Anat. **286**: 571, 1932.
Messina, R.: Folia med. **17**:1225, 1931.
Moehlig, R. C.: Endocrinology **14**:337, 1930.
——and Osius, E. A.: Ann. Int. Med. **4**:579, 1930.
Mönckeberg, J. G.: Virchows Arch. f. path. Anat. **167**:191, 1902.
Moll, T.; Domagk, G., and Laquer, F.: Klin. Wchnschr. **12**:465, 1933.
Montgomery, H.: Proc. Staff Meet., Mayo Clin. **12**: 641, 1937.

Moon, V. H.: Arch. Path. **3**:404, 1927.
Morawitz, P., and Pratt, J.: München. med. Wchnschr. **55**:1817, 1908.
Moschcowitz, E.: Vascular Sclerosis, New York, Oxford University Press, 1942.
Mosebach, W.: Virchows Arch. f. path. Anat. **289**: 646, 1933.
Moxon: Tr. Path. Soc. London **24**:129, 1873.
Müller, C.: Acta med. Scandinav. **89**:75, 1938; Nord. med. **2**:1183, 1939; Arch. Int. Med. **64**:675, 1939.
Müller, O., and Inada, R.: Deutsche med. Wchnschr. **30**:1751, 1904.
Muller, G. L.: Medicine **9**:119, 1930.
Murata, M., and Kataoka, S.: Verhandl. d. jap. path. Gesellsch. **7**:27, 1917; **8**:221, 1918.
Murchison: Tr. Path. Soc. London **20**:187, 1869.
Natali, C.: Frankfurt. Ztschr. f. Path. **47**:1, 1934.
Nékám, L., and Ottenstein, B.: Klin. Wchnschr. **14**: 641, 1935.
Nelson, M. G.: J. Path. & Bact. **53**:105, 1941.
Newburgh, L. H., and Clarkson, S.: J. A. M. A. **79**:1106, 1922; Arch. Int. Med. **31**:653, 1923.
Nikulin, M., and Hetmann, Z.: Arch. f. Gewerbepath. u. Gewerbehyg. **4**:653, 1933.
Nisi, H.: Tr. Soc. path. jap. **29**:272, 1939.
Nuzum, F. R.; Seegal, B.; Garland, R., and Osborne, M.: Arch. Int. Med. **37**:733, 1926.
Nyboer, J.; Bruger, M., and Rabson, S. M.: Am. Heart J. **21**:657, 1941.
Okey, R., and Greaves, V. D.: J. Biol. Chem. **129**: 111, 1939.
——and Stewart, D.: ibid. **99**:717, 1933.
Okuneff, N.: Beitr. z. path. Anat. u. z. allg. Path. **71**:99, 1922-1923.
Onizawa, J.: J. Biochem. **10**:425, 1939.
Ophüls, W.: The Pathogenesis of Arteriosclerosis, in Cowdry, E. V.: Arteriosclerosis, New York, The Macmillan Company, 1933.
Pachorukow, D.: Arb. d. pharmakol. Inst. zu Dorpat **1**:1, 1888.
Page, I. H.: Ann. Int. Med. **14**:1741, 1941.
——and Bernhard, W. G.: Arch. Path. **19**:530, 1935.
——and Farr, L. E.: J. Clin. Investigation **15**:181, 1936.
——and Menschick, W.: Naturwissenschaften **18**:585, 1930.
——Kirby, E., and Van Slyke, D. D.: J. Clin. Investigation **15**:101, 1936.
Pagel, W.: Virchows Arch. f. path. Anat. **258**:416, 1925.
Paterson, J. C.: Arch. Path. **22**:313, 1936.
Pekelharing, C. A.: Beitr. z. path. Anat. u. z. allg. Path. **8**:245, 1890.
Peters, J. P., and Man, E. B.: J. Clin. Investigation **22**:715 and 721, 1943.
Petersilie, H.: J. Lab. & Clin. Med. **20**:144, 1934.
Pick, E. P., and Pineles, F.: Ztschr. f. exper. Path. u. Therap. **7**:518, 1909.
Pick, L.: Deutsche med. Wchnschr. **37**:1930, 1978, 2036 and 2086, 1911.
Pilgersdorfer, W.: Klin. Wchnschr. **19**:512, 1940.
Pineles, F.: Mitt. a. d. Grenzgeb. d. Med. u. Chir. **14**:120, 1904-1905.
Poensgen, A.: Virchows Arch. f. path. Anat. **91**:350, 1883.
Polano, M. K.: Arch. f. Dermat. u. Syph. **174**:213, 1936.
Popken, C.: Beitr. z. path. Anat. u. z. allg. Path. **97**:337, 1936.
Pye-Smith, P. H.: Tr. Path. Soc. London **24**:250, 1873; Guy's Hosp. Rep. **22**:97, 1877.
Raab, W.: München. med. Wchnschr. **86**:689, 1939.
——and Friedmann, R.: Klin. Wchnschr. **16**:1159, 1936.
Rabinowitch, I. M.: Ann. Int. Med. **8**:1436, 1935.
——Richie, W. I., and McKee, S. H.: ibid. **7**:1478, 1934.
Rabl, R.: Virchows Arch. f. path. Anat. **266**:133, 1927-1928.
Radwin, L. S.: Am. J. Dis. Child. **60**:1120, 1940.
Ranke: Beitr. z. path. Anat. u. z. allg. Path. **71**:78, 1923; **75**:269, 1926.
Redfield, R. L.: J. Am. Inst. Homeop. **33**:584, 1940.
Reineck, H.: Beitr. z. path. Anat. u. z. allg. Path. **80**:145, 1928.
Remesow: Verhandl. d. deutsch. path. Gesellsch. **37**: 142, 1934; Zentralbl. f. allg. Path. u. path. Anat. **49**: 361, 1930.
——and Tavaststyerna, N.: Ztschr. f. d. ges. exper. Med. **76**:419, 1931.
Rémond, A.; Colombiés, H., and Bernardbeig, J.: Compt. rend. Soc. de biol. **91**:445, 1924.
Richter, C. P., and Clisby, K. H.: Proc. Soc. Exper. Biol. & Med. **48**:684, 1941.
Roemer, R.: Ztschr. f. Geburtsh. **71**:350, 1912.
Rössle: Kor.-Bl. d. allg. ärztl. Ver. v. Thüringen **49**: 21, 1920.
Rohrschneider, W.: Virchows Arch. f. path. Anat. **256**:139, 1925.
Romberg, E.: Verhandl. d. Kong. inn. Med. **21**:90, 1904; Lehrbuch der Krankheiten des Herzens und der Blutgefässe, Stuttgart, F. Enke, 1921.
Root, H. F.; Bland, E. F.; Gordon, W. H., and White, P. D.: J. A. M. A. **113**:27, 1939.
Rosenthal, F., and Meier, K.: Arch. f. d. ges. exper. Path. **91**:246, 1921.
——Friedländer, E., and Kohn, R.: Arch. f. exper. Path. u. Pharmakol. **175**:343, 1934.
Rosenthal, S. R.: Arch. Path. **18**:473, 660 and 827, 1934.
Sakai, S.: Biochem. Ztschr. **62**:387, 1914.
Sakurai, S.: Jap. J. Exper. Med. **7**:449, 1929,
Saltykow, S.: Verhandl. d. deutsch. path. Gesellsch. **14**:119, 1910; **21**:398, 1926; Virchows Arch. f. path. Anat. **213**:8, 1913; Beitr. z. path. Anat. u. z. allg. Path. **57**:415, 1913.
Sanders: Am. J. M. Sc. **142**:727, 1911.
Sappington, S. W., and Fisher, H. R.: Arch. Path. **34**:989, 1942.
——and Horneff, J. A.: Am. J. M. Sc. **201**:862, 1941.
Saxton, J. A.: New York State J. Med. **41**:1095, 1941.
Scarff, R. W.: J. Path. & Bact. **30**:647, 1927.
Schally, A. O.: Ztschr. f. klin. Med. **128**:376, 1935.
Scheel, O.: Virchows Arch. f. path. Anat. **191**:135, 1908.
Schlossman, N. C., and Gerber, L.: Ann. Surg. **115**: 292, 1942.
Schmetz: Klin. Wchnschr. **15**:805, 1936.
Schmidt, L. H., and Hughes, H. B.: Endocrinology **22**:474, 1938.
Schmidtmann, M.: Virchows Arch. f. path. Anat. **237**:1, 1922; **255**:206, 1925; Verhandl. d. deutsch. path. Gesellsch. **22**:226, 1927.
——and Hüttich, M.: Virchows Arch. f. path. Anat. **267**:601, 1928.
Schönheimer, R.: Ztschr. f. physiol. Chem. **160**:61, 1926; **177**:143, 1928; **211**:65, 1932; Klin. Wchnschr. **43**:1793, 1932; Virchows Arch. f. path. Anat. **249**:1, 1924; **251**:732, 1924.
——and Yuasa, D.: Verhandl. d. deutsch. path. Gesellsch. **25**:304, 1927.
Schottmüller, H.: Klin. Wchnschr. **10**:620, 1931.

Schulte, E.: Beitr. z. path. Anat. u. z. allg. Path. **61**: 570, 1915-1916.

Schultz, A.: Virchows Arch. f. path. Anat. **239**:415, 1922; Verhandl. d. deutsch. path. Gesellsch. **20**:120, 1925.

Scupham, G. W.; de Takáts, G.; Van Dellen, T. R., and Beck, W. C.: Arch. Int. Med. **64**:590, 1939.

Seel, H., and Creuzberg, G.: Arch. f. exper. Path. u. Pharmakol. **161**:674, 1931.

Seemann, G.: Beitr. z. path. Anat. u. z. allg. Path. **83**:705, 1930.

Shapiro, S.: M. J. & Rec. **126**:284, 1927; J. Exper. Med. **45**:595, 1927; Endocrinology **11**:279, 1927.

——and Seecoff, D. P.: J. Lab. & Clin. Med. **10**:826, 1925.

Shelton, E. K.: J. A. M. A. **117**:1948, 1941.

Shillito, F. H.; Bidwell, E. H., and Turner, K. B.: J. Biol. Chem. **112**:551, 1936.

Siegmund: Verhandl. d. deutsch. path. Gesellsch. **18**: 59, 1921.

Simnitzky, S.: Ztschr. f. Heilk. **24**:177, 1903.

Smetana, H.: Virchows Arch. f. path. Anat. **274**:170, 1929; **274**:697, 1930.

Smith, W. F.: Tr. Path. Soc. London **28**:236, 1877.

Sobotka, H.: Macromolecular Layers: Their Application in Physiology and Medicine, in Glasser, O.: Medical Physics, Chicago, The Year Book Publishers, Inc., 1944, p. 763.

Sperry, W. M., in Luck, J. M.: Annual Review of Biochemistry, Stanford University, Calif., Annual Reviews, Inc., 1939, vol. 8, p. 231.

——and Stoyanoff, V. A.: J. Nutrition **9**:131 and 157, 1935.

Ssokoloff, N. A.: Virchows Arch. f. path. Anat. **245**: 203, 1923.

Ssolojew, A.: Virchows Arch. f. path. Anat. **241**:1, 1923; **250**:360, 1924; **261**:253, 1926; **283**:213, 1932; Zentralbl. f. allg. Path. u. path. Anat. **53**: 145, 1932.

Steinbiss, W.: Virchows Arch. f. path. Anat. **212**:152, 1913.

Steiner, A.: Proc. Soc. Exper. Biol. & Med. **38**:231, 1938; **39**:411, 1938.

——and Domanski, B.: Am. J. M. Sc. **201**:820, 1941; **204**:79, 1942; Arch. Int. Med. **71**:397, 1943; Proc. Soc. Exper. Biol. & Med. **55**:236, 1944.

Stepp, W.: Beitr. z. path. Anat. u. z. allg. Path. **69**: 233, 1921.

Stern, R.: Arch. f. exper. Path. u. Pharmakol. **112**: 129, 1926.

Stewart, H. J., and Evans, W. F.: Am. Heart J. **23**: 175, 1942.

Stockman and Charteris: Brit. M. J. **2**:1520, 1901.

Stokes, E. H.: A Clinical and Experimental Investigation of the Blood Cholesterol Content in Myxoedema and Other Conditions, Sydney, Australasian Medical Publishing Co., Ltd., 1941.

Strauss, H.: Acta med. Scandinav. **93**:526, 1937.

Strauss, L. H.: Ztschr. f. exper. Med. **98**:603, 1936.

——and Scheer, P.: Klin. Wchnschr. **15**:187, 1936.

Strome, F. P., and Blaine, B. C.: Pennsylvania M. J. **43**:431, 1940.

Strong, R. A.: J. A. M. A. **107**:422, 1936.

Stukkeĭ, N. V.: Alterations in the Aorta of Rabbits Under the Influence of Excessive Animal Diet: Dietetic Arteriosclerosis, Inaug. Dissert., St. Petersburg, 1910; Zentralbl. f. allg. Path. u. path. Anat. **21**:668, 1910; **22**:379, 1911; **23**:910, 1912.

Stumpf: Beitr. z. path. Anat. u. z. allg. Path. **59**:396, 1914.

Sumikawa, T.: Virchows Arch. f. path. Anat. **196**: 232, 1909.

Svendsen, M.: Acta med. Scandinav. **104**:235, 1940.

Tannenberg, J.: Blutgefässe, in Lichtwitz, L.; Liesegang, R. E., and Spiro, K.: Medizinische Kolloidlehre, Leipzig, Theodor Steinkopff, 1935, p. 570.

Thannhauser, S. J.: Verhandl. d. deutsch. path. Gesellsch. **20**:5, 1925; Lipoidoses: Diseases of the Cellular Lipid Metabolism, New York, Oxford University Press, 1940.

——and Magendantz, H.: Ann. Int. Med. **11**:1662, 1938.

Thiersch. H.: Beitr. z. path. Anat. u. z. allg. Path. **97**:81, 1936.

Thölldte, M.: Beitr. z. path. Anat. u. z. allg. Path. **77**:61, 1927.

Thomas, W. A.: J. A. M. A. **88**:1559, 1927.

Thompson, K. W., and Long, C. N. H.: Endocrinology **28**:715, 1941.

Thompson, W. O.: J. A. M. A. **117**:441, 1941.

Tregubow, A.: Krankheitsforsch. **6**:87, 1928.

Tropp, C.: Klin. Wchnschr. **15**:562, 1936.

Turner, K. B.: J. Exper. Med. **58**:115, 1933.

——and Bidwell, E. H.: ibid. **62**:721, 1935.

——and Khayat, G. B.: ibid. **58**:127, 1933.

——and Steiner, A.: J. Clin. Investigation **18**:45, 1939.

——Present, C. H., and Bidwell, E. H.: J. Exper. Med. **67**:111, 1938.

Uchiyama, J.: Virchows Arch. f. path. Anat. **277**: 642, 1930.

Ungar, H.: Arch. f. exper. Path. u. Pharmakol. **175**: 536, 1934.

Vander Veer, J. B.: M. Clin. North America **23**:1561, 1939.

Vermeulen, C. W.; Allen, J. G.; Clark, D. E.; Julian, O. C., and Dragstedt, L. R.: Proc. Am. Physiol. Soc., 1941.

——Dragstedt, L. R.; Clark, D. E.; Julian, O. C., and Allen, J. G.: Arch. Surg. **42**:260, 1942.

Versé, M.: Beitr. z. path. Anat. u. z. allg. Path. **63**: 780. 1917; Verhandl. d. deutsch. path. Gesellsch. **19**:163, 1923; **20**:67, 1925; Virchows Arch. f. path. Anat. **250**:252, 1924; Zentralbl. f. allg. Path. u. path. Anat. **34**:614, 1924.

Virchow, R.: Phlogose und Thrombose im Gefässsystem, in Gesammelte Abhandlungen zur wissenschaftlichen Medizin, Frankf. a. M., Meidinger Sohn & Co., 1856.

Volhard, F.: Die doppelseitigen hämatogenen Nierenerkrankungen, in Mohr, L., and Staehelin, R.: Handbuch der inneren Medizin, ed. 2, Berlin, Julius Springer, 1931, vol. 6, pt. 1, p. 1.

Wacker, L., and Hueck, W.: München. med. Wchnschr. **60**:2097, 1913; Arch. f. exper. Path. u. Pharmakol. **71**:373, 1912-1913; **74**:416, 1913.

Wada, K.: Tr. Jap. Path. Soc. **16**:179, 1926.

Wail, S. S.: Virchows Arch. f. path. Anat. **245**:219, 1923.

Warischtew, W. K.: Inaug. Dissert., Warsaw, 1914.

Warren, S.: The Pathology of Diabetes Mellitus, ed. 2, Philadelphia, Lea & Febiger, 1938, p. 246.

Wartman, W. B.: Factors Concerned in Narrowing or Occlusion of Coronary Vessels, in Blood, Heart and Circulation, Washington, D. C., American Association for the Advancement of Science, 1940, vol. 13, p. 122.

Weber, F. R.: Cutaneous Xanthoma and Xanthomatosis of Other Parts of the Body: Pituitary Xanthomatosis — Xanthomyelomata of Tendon Sheaths, etc. and the "Cholesterin-Diathesis," London, H. K. Lewis & Company, 1924.

Wegelin: Virchows Arch. f. path. Anat. **254**:689, 1925; Schilddrüse, in Henke, F., and Lubarsch, O.: Handbuch der speziellen Anatomie und Histologie, Berlin, Julius Springer, 1925, vol. 8, pt. 1.

Weinhouse, S.: Arch. Path. **35**:438, 1943.

——and Hirsch, E. F.: ibid. **29**:31, 1940; **30**:856, 1940.

Wepler, W.: Virchows Arch. f. path. Anat. **295**:546, 1935.

Wesselkin, N. W.: Virchows Arch. f. path. Anat. **212**:225, 1913.

Westphal, K.: Klin. Wchnschr. **5**:1995, 1926; Ztschr. f. klin. Med. **101**:545, 558, 584 and 566, 1924-1925.

Westra, J. J., and Kunde, M. M.: Am. J. Physiol. **103**:1, 1933.

White, P. D.: Diabetes in Childhood and Adolescence, Philadelphia, Lea & Febiger, 1932, p. 178; J. A. M. A. **113**:27, 1939.

Whitehead, V. I. E.: Brit. J. Exper. Path. **24**:192, 1943.

Wilens, S. L.: Am. J. Path. **19**:293, 1943.

Wilkins, L.; Fleischmann, W., and Block, W.: J. Clin. Endocrinol. **1**:3 and 91, 1941; J. A. M. A. **116**:2459, 1941.

Winternitz, M. C.: Views as to Causes of Coronary Sclerosis, in Blood, Heart and Circulation, Washington, D. C., American Association for the Advancement of Science, 1940, vol. 13, p. 114; Arch. Path. **28**:124, 1939.

——Thomas, R. M., and LeCompte, P. M.: The Etiology of Arteriosclerosis, Springfield, Ill., Charles C Thomas, Publisher, 1938.

Wise, F.: Arch. Dermat. & Syph. **47**:739, 1943.

Wislicki, L.: Klin. Wchnschr. **8**:1568, 1929.

Wolff, K.: Virchows Arch. f. path. Anat. **293**:472, 1934.

Wolkoff, K.: Beitr. z. path. Anat. u. z. allg. Path. **85**:386, 1930.

Wright, I.: Arch. Surg. **40**:163, 1940.

Yuasa, D.: Beitr. z. path. Anat. u. z. allg. Path. **80**: 570, 1928.

Zeek, P. M.: Am. J. Path. **12**:115, 1936.

Zinserling, W. D.: Beitr. z. path. Anat. u. z. allg. Path. **71**:292, 1923; Virchows Arch. f. path. Anat. **255**:677, 1925; Zentralbl. f. allg. Path. u. path. Anat. **24**:627, 1913; **32**:534, 1921-1922.

Zon, L.: Arch. Path. **27**:888, 1939.

Zondek, H.: Lancet **2**:310, 1941.

COLLOIDAL PLASMATIC DISTURBANCES: C. PROTEINIC TYPE

Disturbances of the quantitative and qualitative composition of the plasma proteins are present in a great number of different diseases (myelomatosis, hyperfibrinogenemia, amyloidosis, immune reactions, chemoallergies, hemoglobinemia, nephrosis, leukemia, hepatic cirrhosis, myxedema, venereal lymphogranuloma, kala-azar) and are artificially produced by the intravenous introduction of homologous or heterologous protein solutions (plasma proteins, gelatin, isinglass, ovalbumin, azoproteins) or by the formation of protein complexes with aromatic substances (sulfonamide compounds, Evans blue). Such disturbances may result from shifts in the ratio of the various plasma protein fractions without causing any increase in the total amount of plasma proteins, or from absolute increases or decreases of one fraction, or from the formation of abnormal endogenous proteins (paraproteins) of smaller or larger size than the ordinary ones, or from combination of a normal plasma protein with an endogenous or exogenous chemical complex, or from the introduction of exogenous proteins. Such changes elicit definite alterations in the colloidal equilibrium of the plasmatic proteins and modify their stability in solution and their reactivity to each other and to the proteins contained in the tissues and the tissue fluids.

Among the three normal plasma proteins (fibrinogen, globulin, albumin) which compose about 6 to 7 per cent of the plasma, albumin has the smallest molecular weight (about 70,000) and the most uniform and symmetric molecular shape, exerts the greater part of the total colloidal osmotic pressure of the plasma, is least readily denatured, is most finely dispersed, has the largest electric charge, the lowest viscosity, the greatest solubility and stability in solution and represents over 60 per cent of the total plasma protein. Globulin, which like albumin normally has a round-shaped molecule, has a molecular weight of about 120,000, is more asymmetrically built, more labile in solution, more viscous, exerts less colloidal osmotic pressure and occurs in three varieties, alpha, beta and gamma. Fibrinogen has a fibrillar molecule, represents about 5 per cent of the total plasma protein and has the largest molecular weight. Alpha globulin combines with certain carbohydrate groups to form mucoglobulins (Cohn). The plasma proteins occur in the plasma in a hydrated form; i. e., by adsorption or hydration they have taken up water and thus have increased their molecular size considerably (Bennhold). The degree and the type of colloidal dispersion of the different plasma proteins may vary considerably, depending on the associated physicochemical conditions of the plasma, and are not reflected by the albumin-globulin ratio (Melnick).

In the hypoproteinemia characterizing lipoid nephrosis fibrinogen and alpha and beta globulins are increased, while gamma globulin and albumin are decreased. In febrile conditions mainly alpha globulin is increased. Immune reactions are associated with hyperproteinemia caused particularly by increases in gamma globulin. Some antibody globulins (horse, pig and cow antipneumococcus euglobulins) may assume enormous sizes through polymerization of smaller units (molecular weight of about 900,000) and exist close to their isoelectric point (Heidelberger). Antibody globulins belong to the gamma variety, from which they may differ by the way in which the polypeptide chain is refolded or recoiled

(Pauling). Hyperproteinemia of immune or other genesis is associated with increased plasma viscosity and accelerated erythrocytic sedimentation and, in severe cases, with pseudoagglutination of erythrocytes (Vignati and Rauchenberg; Foord and Randall; Jeghers and Selesnick; Kracke and Hoffman).

Myelomatosis.—The most striking example of plasmatic proteinic imbalances is found in the quantitative and qualitative abnormalities of the plasma proteins occurring in myelomatosis (Shirer, Duncan and Haden; Jacobson; Cantarow; Kagan). Bing stated that myelomatosis is the most frequent cause of severe hyperproteinemia. Schumacher, Williams and Coltrin observed hyperproteinemia in 23.39 per cent of their cases of myelomatosis; Feller and Fowler, in 9 of 52 cases. According to physicochemical and immunologic studies (Kabat, Moore and Gutman; Gutman, Moore, Gutman, McClellan and Kabat; and Kekwick), three different types of myelomatosis can be distinguished on the basis of the patterns of the plasma protein changes. In one type the gamma globulins are mainly increased, in the second type the beta globulins are elevated, while in the third a normal ratio prevails.

In addition to these quantitative deviations in the total plasma proteins and their individual normal fractions, there occurs in myelomatosis in 65 to 80 per cent of the cases (Geschickter and Copeland; Magnus-Levy; Apitz) an abnormal protein, the Bence Jones protein, which coagulates when heated to 65 C., has a molecular weight of about 35,000 to 40,000 and may combine chemically with pseudoglobulins and other proteins (Mahle, Seed and Welker). The investigations of Bayne-Jones and Wilson and of Robinson have shown that this abnormal endogenous protein is antigenic. It is excreted with the urine and may form casts in the renal tubules by being precipitated out of colloidal mixtures by coacervation. Protein crystals are not infrequently found in the renal tissue (Abrikosoff and Wulff; Gunn and Mahle).

The third type of proteinic plasmatic abnormality occurring in myelomatosis is represented by the occasional appearance of a hypermacromolecular, highly viscous globulin of a molecular weight between 162,000 and 200,000 (von Bonsdorff, Groth and Packalén; Shapiro, Ross and Moore; Perlzweig, Delrue and Geschickter; Bing; Wintrobe and Buell). This protein on chilling settles out from the plasma in the lower layer and when dropped into distilled water precipitates in floccules. When heated to 71 C. it coagulates. Its presence in the blood is usually associated with circulatory disturbances. Bell found a highly viscous globulin plugging the glomerular capillaries in cases of myelomatosis. Perlzweig, Delrue and Geschickter proposed that the hyperproteinemia associated with myelomatosis may possibly be a systemic response to protracted intoxication with a foreign protein, considering Bence Jones protein as a foreign protein. In the light of these findings it is considered significant that myelomatosis is often associated with the occurrence of a proteinosis, amyloidosis (Volland; Apitz; Rosenblum and Kirshbaum; Magnus-Levy; Chester; Rosenblatt; Rosenheim and Wright; Randall).

Noninflammatory Vascular Reactions: (*a*) Hyalinosis: Three types of interstitial or conjunctival proteinic deposits are encountered in vascular walls, namely, amyloid, hyaline and fibrinoid. There are definite relations between these three substances according to their behavior with certain metachromatic stains (Letterer; Dietrich). Loeschke considered interstitial hyaline substance as a product of precipitated protein which has a globulin-like character (Müller), becomes adsorbed to collagen fibrils and shares with amyloid and fibrinoid the quality that it is the result of a deposition of concentrated protein gels (Dietrich) which infiltrated as sols from the blood (Anitschkow). Loeschke proposed that in arteriosclerosis a precipitin protein is deposited on the endothelium which is formed by the interaction of an antigenic protein with an antibody globulin, combining to an insoluble protein and precipitated as a hyaline substance. Amyloid represents, according to Loeschke, only a special variety of hyaline substance. If the antibody formation predominates under such circumstances the site of protein precipitation is near the location where the antibodies are produced. When, on the other hand, the antigen predominates, the site of protein precipitation in the tissues where the antigen is elaborated or released is of minor magnitude, as the antigen enters the blood and precipitation then occurs everywhere in the vascular system. If there is an overwhelming predominance of the amount of antigen over the quantity of antibodies available, the precipitation of the protein complex takes place mainly at the site of the antibody production in the spleen and the reticuloendothelial system. Hyalin is formed at places where arteriosclerotic nutritive disturbances cause a degradation of tissues. As lipoids are often in some way bound to protein, they are precipitated with the hyalin and thus become embedded into the hyaline and amyloid matter. While a part of the hyaline deposits in arteriosclerotic vascular walls may be attributed to a summation of the deposi-

tion of hyaline matter occurring during the course of life as a reaction to various diseases passed through, another part of the hyaline substance may be generated in response to specific factors, probably representing proteinic constituents of the blood.

The resemblance of hyaline and amyloid vascular deposits is shown by their reactivity to certain stains and by their local distribution in the vascular walls. Both substances occur in noninflammatory, chronic degenerative types of vascular lesions. These substances, as well as the processes leading to their deposition, therefore lack irritative and toxic properties. The vascular lesions elicited by them are similar to those caused by other nontoxic proteins, such as gelatin and ovalbumin, and differ markedly from the inflammatory vascular reactions in which fibrinoid deposits are observed and in which more or less acute necrotizing toxic influences prevail. Immune or allergic reactions play apparently a significant role in the production of both types of lesions and their underlying or associated plasmatic colloidal proteinic disturbances.

(*b*) Amyloidosis: The development of the proteinosis amyloidosis not only is characterized by the presence of certain hematologic reactions (anemia, acceleration of erythrocytic sedimentation, increase in viscosity of serum) but is accompanied by more or less marked hyperproteinemia and especially hyperglobulinemia, which may become occasionally so excessive that the globulin portion of the plasma will separate from the nonglobulin part on standing (Bing; Eklund and Reimann; Reimann, Koucky and Eklund; Magnus-Levy; Morgenstern). This disease, which usually appears in man in connection with chronic suppurative conditions or disorders associated with protein wastage or with disturbances of protein metabolism (osteomyelitis, chronic tuberculosis, syphilis, lymphogranuloma, myeloma), leads to extracellular and intracellular deposition of a complex proteinic substance (amyloid) (Cohen; Danisch; Willer), especially in and around vascular and capillary walls (Hass and Schülz). As amyloidosis has been produced experimentally in animals by repeated and protracted parenteral introduction of certain foreign proteins—for example, in horses used for the production of antiserum (Arndt; Doerken; Reitstötter), rabbits, mice and others—or by feeding an unnatural proteinic diet (Jaffé; Grayzel, Jacobi, Marshall, Bogin and Bolker; Kuczinski; Ku and Simon, and others), amyloid has been related to excessive formation of a normal or an abnormal antigen-antibody complex (Letterer; Loeschke; Primgaard; Lucké and Markley; Koletsky and Stecher) or has been considered to be the result of a reaction of hypersensitivity.

Amyloidosis has been seen in man also in connection with chronic poisonings by lead, manganese and alcohol (Celli; Butt; Israel). It may be mentioned in this connection that the investigations of Ehrström have shown that a mixture of serum with chondroitin–sulfuric acid apparently precipitates albumin, causing thereby a relative increase of the serum globulin fraction. Murata and Yoshikawa observed that amyloid is deposited in rabbits given injections of, or fed, silicic acid, at sites where the silica accumulates in the tissues and undergoes a change from a sol into a gel, thereby showing that the physicochemical conditions (p_H) of the tissue play a significant role in the distribution of amyloid (Morgenstern). Regardless of the primary or the secondary nature of amyloid, this proteinic matter is always deposited in the wall of the small and the medium-sized arteries of the various organs (heart, kidney, tongue and other organs) (Strauss; Binford; Koletzky and Stecher; Pearson, Rice and Dickens; Bell; Perla and Gross; Dillon and Evans). In some cases the aorta is also involved by medial nodular amyloid deposits. This proteinic condensation product, which usually contains 5 to 6 per cent of cholesterol, resembling therein the arteriolar subendothelial hyaline deposits with their lipoidal content, involves the subendothelial space and the media, where it spreads in between the muscle cells in homogeneous branching structures which ultimately merge into a solid mass under atrophy of the muscular elements (Peters). In large arteries the amyloid occupies in general the outer part of the media and the adventitia, whence it extends into the vascular wall along the amyloidotic vasa vasorum. The walls of the smaller vessels are highly thickened, and their lumens are usually considerably narrowed.

While M. B. Schmidt and also Leupold claimed that a precursor of amyloid circulates in the blood and penetrates from there into the vascular walls, Letterer suspected that the preamyloid substance is formed extravascularly and is precipitated in the vascular walls while passing through these tissues on its way to the blood.

Dick and Leiter observed in 12 per cent of rabbits given injections of various bacterial cultures medial necroses, calcifications and atheromatosis of the aorta in addition to widespread amyloidosis. This incidence represents, according to these investigators, six times the normal incidence of aortic lesions.

Hueper recently produced in dogs vascular lesions that were comparable in some respects by

injecting large amounts of solutions of foreign proteins, namely, gelatin and ovalbumin, intravenously over long periods. Dogs given solutions of ovalbumin or of an aminoantipyrine azocompound of ovalbumin had in the intima of the aorta and its branches hydropic mononuclear cell infiltrations, fibroblastic loose cushions and hyaline thickenings and in the media focal hyalinization and calcification. Some of the renal arterioles and small arteries exhibited thickened and locally hyalinized walls with proliferation of the endothelium.

The aortas of the dogs given solutions of gelatin, which elicits in the blood pseudoagglutination of the erythrocytes (Parkins, Koop, Riegel, Vars and Lockwood) and behaves in some respects like globulin (Brunschwig, Scott, Corbin and Moe), displayed extensive edema and hyalinization of the intima with fine granular calcium deposits in the thickenings and in the internal elastic membranes, as well as large hyalinizations and marked edema of the media. The walls of the renal arteries and arterioles were vacuolated and edematous, and medial hyalinization and intimal hyaline thickenings were found in some instances (Hueper). Some of the arterioles were occluded by hyaline masses infiltrating the walls and containing irregularly arranged and shaped nuclei.

In this connection brief mention may be made of an observation recorded during the course of a toxicopathologic study of animals given injections of Evans blue (Hueper and Ichniowski). Eight molecules of this dye combine in the blood with one molecule of albumin, forming thereby a complex having the physicochemical characteristics of globulin (Rawson; Gregersen and Rawson). The dye is retained in the aortic wall and in other organs, particularly the testes, the teeth and the cartilage, over periods of more than six months. The aortas of dogs thus treated revealed small foci of intimal hyalinization, while the renal glomeruli of rabbits exhibited extensive hyalinizations. However, it is still uncertain whether or not the disturbance in the character of the serum albumins has any causal relation to these vascular manifestations. A similar binding of sulfonamide compounds to plasma albumins (Davis; Schonholzer) may be involved in the appearance of medial hyaline degenerations and calcifications of the aorta and the pulmonary and coronary arteries of rats following the administration of these chemicals (Endicott, Kornberg and Daft; Lehr, Antopol, Churg, and Spring; Daft, Ashburn, Spicer and Sebrell; Ashburn, Daft, Endicott and Sebrell). It is problematic whether or not extensive calcinotic degenerative aortic lesions in mice painted with methylcholanthrene and fed a cystine-deficient diet are of similar genesis (White and Mider; White, Mider and Heston), or whether nutritional or vitamin deficiencies or metabolic disturbances play a causal role.

COLLOID PLASMATIC DISTURBANCES:
C. PROTEINIC TYPE

Noninflammatory Vascular Reactions:

Abrikosoff, A., and Wulff, F.: Verhandl. d. deutsch. path. Gesellsch. **22**:270, 1927.

Anitschkow, N.: Klin. Wchnschr. **4**:2233, 1925.

Apitz, K.: Klin. Wchnschr. **19**:1058, 1940; Virchows Arch. f. path. Anat. **306**:630, 1940.

Arndt, H. J.: Arch. f. Tierh. **63**:1, 1931; Klin. Wchnschr. **10**:910, 1931; Verhandl. d. deutsch. path. Gesellsch. **26**:243, 1931.

Artificial Antibodies, editorial, Science **96**:181, 1942.

Ashburn, L. L.; Daft, F. S.; Endicott, K. M., and Sebrell, W. H.: Pub. Health Rep. **57**:1883, 1942.

Askanazy, M.: Verhandl. d. deutsch. path. Gesellsch. **7**:32, 1904.

Bell, E. T.: Am. J. Path. **9**:185, 1933.

Beunhold, H.: Klin. Wchnschr. **11**:2057, 1932.

Binford, C. H.: Arch. Path. **29**:314, 1940.

Bing, J.: Acta med. Scandinav. **103**:547, 1940.

von Bonsdorff, B.; Groth, H., and Packalén, T.: Acta med. Scandinav., 1938, supp. 89, p. 347.

Brunschwig, A.; Scott, V. B.; Corbin, N., and Moe, R.: Proc. Soc. Exper. Biol. & Med. **52**:46, 1943.

Butt, E. M.: Arch. Path. **10**:859, 1930.

Cantarow, A.: Am. J. M. Sc. **189**:425, 1935.

Celli, P.: Sperimentale, Arch. di biol. **89**:749, 1935; abstracted, Zentralbl. f. allg. Path. u. path. Anat. **65**:388, 1936.

Chester, W.: Ztschr. f. klin. Med. **124**:466, 1923.

Cohen, S.: Ann. Int. Med. **19**:990, 1943.

Cohn, E. J.: Chem. Rev. **28**:395, 1941.

Daft, F. S.; Ashburn, L. L., and Sebrell, W. H.: Science **96**:321, 1942.

——Ashburn, L. L.; Spicer, S. S., and Sebrell, W. H.: Pub. Health Rep. **57**:217, 1942.

Danisch: Verhandl. d. deutsch. path. Gesellsch. **20**:307, 1925.

Davis, B. D.: Science **95**:78, 1942.

Dick, G. F., and Leiter, L.: Am. J. Path. **17**:741, 1941.

Dietrich, A.: Verhandl. d. deutsch. path. Gesellsch. **21**: 156, 1926.

Dillon, J. A., and Evans, L. R.: Ann. Int. Med. **17**: 722, 1942.

Doerken, E.: Virchows Arch. f. path. Anat. **286**:487, 1932.

Ehrström, M. C.: Acta med. Scandinav. **101**:551, 1939.

Eklund, C. M., and Reimann, H. A.: Arch. Path. **21**:1, 1936.

Endicott, K. M.; Kornberg, A., and Daft, F. S.: Pub. Health Rep. **59**:49, 1944.

Feller, A. E., and Fowler, W. M.: J. Lab. & Clin. Med. **23**:369, 1938.

Foord, A. G., and Randall, L.: Am. J. Clin. Path. **5**:532, 1935.

Grayzel, H. G.; Jacobi, M.; Marshall, H. B.; Bogin, M., and Bolker, H.: Arch. Path. **17**:50, 1934.

Gregersen, M. I., and Rawson, R. A.: Am. J. Physiol. **138**:698, 1943.

Gunn, F. D., and Mahle, A. E.: Arch. Path. **26**:377, 1938.

Gutman, A. B.; Moore, D. H.; Gutman, E. B.; McClellan, V., and Kabat, E. A.: J. Clin. Investigation **20**:765, 1941.
Hueper, W. C.: Am. J. Path. **18**:895, 1942.
——and Ichniowski, C. T.: Arch. Surg. **48**:17, 1944.
Israel, I.: Ein Fall von lokalem Amyloid, Inaug. Dissert., Tübingen, Bochum-Langendreer, H. Poppinghaus, 1933.
Jacobson, V. C.: J. Urol. **1**:167, 1917.
Jeghers, H., and Selesnick, S.: Internat. Clin. **47**:248, 1937.
Kabat, E. A.; Moore, D. H., and Gutman, A. B.: Paper presented at the meeting of the American Chemical Society, Buffalo, N. Y., 1942.
Kagan, B. M.: Am. J. M. Sc. **206**:309, 1943.
Kekwick, R. A.: Biochem. J. **34**:1248, 1940.
Koletsky, S., and Stecher, R. M.: Arch. Path. **27**: 267, 1939.
Kracke, R. R., and Hoffman, B. J.: Ann. Int. Med. **19**:673, 1943.
Ku, D. Y., and Simon, M. A.: Arch. Path. **18**:245, 1934.
Letterer, E.: Beitr. z. path. Anat. u. z. allg. Path. **75**: 486, 1926; Verhandl. d. deutsch. path. Gesellsch. **20**:301, 1925.
Leupold, E.: Beitr. z. path. Anat. u. z. allg. Path. **64**: 347, 1917-1918.
Loeschke, H.: Beitr. z. path. Anat. u. z. allg. Path. **77**:231, 1927.
Mahle, A. E.; Seed, L., and Welker, W. H.: Arch. Path. **26**:441, 1938.
Melnick, D.; Field, H., and Parnall, C. G.: Arch. Int. Med. **66**:295, 1940.
Morgenstern, Z.: Virchows Arch. f. path. Anat. **259**: 698, 1926.
Müller, E.: Beitr. z. path. Anat. u. z. allg. Path. **97**:42, 1936.
Murata, M., and Yoshikawa, S.: Virchows Arch. f. path. Anat. **264**:587, 1927.
Parkins, W. M.; Koop, C. E.; Riegel, C.; Vars, H. M., and Lockwood, J. S.: Ann. Surg. **118**:193, 1943.
Pauling, L.: J. Am. Chem. Soc. **62**:2643, 1940.
Pearson, B.; Rice, M., and Dickens, K.: Arch. Path. **32**:1, 1941.
Perla, D., and Gross, H.: Am. J. Path. **11**:93, 1935.
Perlzweig, W. A.; Delrue, G., and Geschickter, C.: J. A. M. A. **90**:755, 1929.
Peters, J. T.: Arch. Path. **35**:832, 1943.
Randall, O. S.: Am. J. Cancer **19**:838, 1933.
Rawson, R. A.: Am. J. Physiol. **138**:706, 1943.
Reimann, H. A.; Koucky, R. F., and Eklund, C. M.: Am. J. Path. **11**:977, 1935.
Rosenblatt, M. B.: Ann. Int. Med. **8**:678, 1934.
Rosenblum, A. H., and Kirshbaum, J. D.: J. A. M. A. **106**:988, 1936.
Rosenheim, M. L., and Wright, G. P.: J. Path. & Bact. **37**:332, 1933.
Schonholzer, G.: Klin. Wchnschr. **19**:790, 1940.
Shapiro, S.; Ross, V., and Moore, D. H.: J. Clin. Investigation **22**:137, 1943.
Shirer, J. W.; Duncan, W., and Haden, R. L.: Arch. Int. Med. **50**:829, 1932.
Strauss, A.: Virchows Arch. f. path. Anat. **291**:219, 1933.
Sweigert, C. F.: Am. J. M. Sc. **190**:246, 1935.
Ulrich, H.: Arch. Int. Med. **64**:994, 1939.
Vignati, J., and Rauchenberg, M.: Klin. Wchnschr. **16**:62, 1937.
Volland, W.: Virchows Arch. f. path. Anat. **298**:660, 1937.
White, J., and Mider, G. B.: J. Nat. Cancer Inst. **2**:95, 1941.
——Mider, G. B., and Heston, W. E.: ibid. **3**:453, 1943.
Willer, H.: Frankfurt. Ztschr. f. Path. **46**:306, 1933.
Wintrobe, M. M., and Buell, M. V.: Bull. Johns Hopkins Hosp. **52**:156, 1933.

Inflammatory Vascular Reactions.—(*a*) Allergy: During the preceding discussions the occurrence of inflammatory, fibrinoid, necrotizing lesions affecting mainly the small and the medium-sized arteries has been repeatedly mentioned in connection with, and as complications of, vasotonic as well as hydrostatic arteriosclerotic changes. It was pointed out on these occasions that apparently superimposed allergic conditions are responsible for this type of reaction. In view of the fact that these manifestations are accompanied by an infiltration of the vascular wall by plasmatic material, giving rise to fibrinoid deposits, it appears likely that the inflammatory and necrotizing variety of vascular change occurs in association with plasmatic colloidal proteinic disturbances of allergic-hyperergic nature when the immune bodies formed during these processes possess toxic properties. The hyperproteinemia occurring during immune conditions is characterized by an increase in fibrinogen and globulin (Schittenhelm) and is associated with colloidoclastic leukopenia, thrombopenia, eosinophilia, a lengthened clotting time and an accelerated rate of sedimentation of the erythrocytes during acute crises of the immune state (allergic or anaphylactic shock). Such reactions are apparently related to the degree of dispersion of the interreacting colloidal particles, as colloids consisting of highly dispersed small particles are less reactive than those with coarse particles (Klopstock).

Numerous observations connect allergic reactions with the development of degenerative arterial disease. Claims to this effect have been advanced repeatedly in regard to various vasospastic conditions (thromboangiitis obliterans, angina pectoris, reactions suggesting Raynaud's disease, malignant hypertension) caused by endogenous or exogenous physical and chemical agents, such as foodstuffs, nicotine, pollen, drugs and cold (Vaughan; Davison, Thoroughman and Bowcock; van Creveld; Werley; Sullivan and Vaughan; Krauspe; von Eiselsberg; Gaensslen; Lichtwitz; Schmidt; Conti; Dattner; Kämmerer). Joyner and Sabin reported that in certain allergic conditions the permeability of the capillary endothelium is reduced, preventing the removal of colloidal nontoxic dyes, while Klinge pointed out that anaphylactic shock can be prevented by the intravenous injection of colloidal

injecting large amounts of solutions of foreign proteins, namely, gelatin and ovalbumin, intravenously over long periods. Dogs given solutions of ovalbumin or of an aminoantipyrine azocompound of ovalbumin had in the intima of the aorta and its branches hydropic mononuclear cell infiltrations, fibroblastic loose cushions and hyaline thickenings and in the media focal hyalinization and calcification. Some of the renal arterioles and small arteries exhibited thickened and locally hyalinized walls with proliferation of the endothelium.

The aortas of the dogs given solutions of gelatin, which elicits in the blood pseudoagglutination of the erythrocytes (Parkins, Koop, Riegel, Vars and Lockwood) and behaves in some respects like globulin (Brunschwig, Scott, Corbin and Moe), displayed extensive edema and hyalinization of the intima with fine granular calcium deposits in the thickenings and in the internal elastic membranes, as well as large hyalinizations and marked edema of the media. The walls of the renal arteries and arterioles were vacuolated and edematous, and medial hyalinization and intimal hyaline thickenings were found in some instances (Hueper). Some of the arterioles were occluded by hyaline masses infiltrating the walls and containing irregularly arranged and shaped nuclei.

In this connection brief mention may be made of an observation recorded during the course of a toxicopathologic study of animals given injections of Evans blue (Hueper and Ichniowski). Eight molecules of this dye combine in the blood with one molecule of albumin, forming thereby a complex having the physicochemical characteristics of globulin (Rawson; Gregersen and Rawson). The dye is retained in the aortic wall and in other organs, particularly the testes, the teeth and the cartilage, over periods of more than six months. The aortas of dogs thus treated revealed small foci of intimal hyalinization, while the renal glomeruli of rabbits exhibited extensive hyalinizations. However, it is still uncertain whether or not the disturbance in the character of the serum albumins has any causal relation to these vascular manifestations. A similar binding of sulfonamide compounds to plasma albumins (Davis; Schonholzer) may be involved in the appearance of medial hyaline degenerations and calcifications of the aorta and the pulmonary and coronary arteries of rats following the administration of these chemicals (Endicott, Kornberg and Daft; Lehr, Antopol, Churg, and Spring; Daft, Ashburn, Spicer and Sebrell; Ashburn, Daft, Endicott and Sebrell). It is problematic whether or not extensive calcinotic degenerative aortic lesions in mice painted with methylcholanthrene and fed a cystine-deficient diet are of similar genesis (White and Mider; White, Mider and Heston), or whether nutritional or vitamin deficiencies or metabolic disturbances play a causal role.

COLLOID PLASMATIC DISTURBANCES:
C. PROTEINIC TYPE

Noninflammatory Vascular Reactions:

Abrikosoff, A., and Wulff, F.: Verhandl. d. deutsch. path. Gesellsch. **22**:270, 1927.

Anitschkow, N.: Klin. Wchnschr. **4**:2233, 1925.

Apitz, K.: Klin. Wchnschr. **19**:1058, 1940; Virchows Arch. f. path. Anat. **306**:630, 1940.

Arndt, H. J.: Arch. f. Tierh. **63**:1, 1931; Klin. Wchnschr. **10**:910, 1931; Verhandl. d. deutsch. path. Gesellsch. **26**:243, 1931.

Artificial Antibodies, editorial, Science **96**:181, 1942.

Ashburn, L. L.; Daft, F. S.; Endicott, K. M., and Sebrell, W. H.: Pub. Health Rep. **57**:1883, 1942.

Askanazy, M.: Verhandl. d. deutsch. path. Gesellsch. **7**:32, 1904.

Bell, E. T.: Am. J. Path. **9**:185, 1933.

Beunhold, H.: Klin. Wchnschr. **11**:2057, 1932.

Binford, C. H.: Arch. Path. **29**:314, 1940.

Bing, J.: Acta med. Scandinav. **103**:547, 1940.

von Bonsdorff, B.; Groth, H., and Packalén, T.: Acta med. Scandinav., 1938, supp. 89, p. 347.

Brunschwig, A.; Scott, V. B.; Corbin, N., and Moe, R.: Proc. Soc. Exper. Biol. & Med. **52**:46, 1943.

Butt, E. M.: Arch. Path. **10**:859, 1930.

Cantarow, A.: Am. J. M. Sc. **189**:425, 1935.

Celli, P.: Sperimentale, Arch. di biol. **89**:749, 1935; abstracted, Zentralbl. f. allg. Path. u. path. Anat. **65**:388, 1936.

Chester, W.: Ztschr. f. klin. Med. **124**:466, 1923.

Cohen, S.: Ann. Int. Med. **19**:990, 1943.

Cohn, E. J.: Chem. Rev. **28**:395, 1941.

Daft, F. S.; Ashburn, L. L., and Sebrell, W. H.: Science **96**:321, 1942.

——Ashburn, L. L.; Spicer, S. S., and Sebrell, W. H.: Pub. Health Rep. **57**:217, 1942.

Danisch: Verhandl. d. deutsch. path. Gesellsch. **20**:307, 1925.

Davis, B. D.: Science **95**:78, 1942.

Dick, G. F., and Leiter, L.: Am. J. Path. **17**:741, 1941.

Dietrich, A.: Verhandl. d. deutsch. path. Gesellsch. **21**: 156, 1926.

Dillon, J. A., and Evans, L. R.: Ann. Int. Med. **17**: 722, 1942.

Doerken, E.: Virchows Arch. f. path. Anat. **286**:487, 1932.

Ehrström, M. C.: Acta med. Scandinav. **101**:551, 1939.

Eklund, C. M., and Reimann, H. A.: Arch. Path. **21**:1, 1936.

Endicott, K. M.; Kornberg, A., and Daft, F. S.: Pub. Health Rep. **59**:49, 1944.

Feller, A. E., and Fowler, W. M.: J. Lab. & Clin. Med. **23**:369, 1938.

Foord, A. G., and Randall, L.: Am. J. Clin. Path. **5**:532, 1935.

Grayzel, H. G.; Jacobi, M.; Marshall, H. B.; Bogin, M., and Bolker, H.: Arch. Path. **17**:50, 1934.

Gregersen, M. I., and Rawson, R. A.: Am. J. Physiol. **138**:698, 1943.

Gunn, F. D., and Mahle, A. E.: Arch. Path. **26**:377, 1938.

Gutman, A. B.; Moore, D. H.; Gutman, E. B.; McClellan, V., and Kabat, E. A.: J. Clin. Investigation **20**:765, 1941.
Hueper, W. C.: Am. J. Path. **18**:895, 1942.
——and Ichniowski, C. T.: Arch. Surg. **48**:17, 1944.
Israel, I.: Ein Fall von lokalem Amyloid, Inaug. Dissert., Tübingen, Bochum-Langendreer, H. Poppinghaus, 1933.
Jacobson, V. C.: J. Urol. **1**:167, 1917.
Jeghers, H., and Selesnick, S.: Internat. Clin. **47**:248, 1937.
Kabat, E. A.; Moore, D. H., and Gutman, A. B.: Paper presented at the meeting of the American Chemical Society, Buffalo, N. Y., 1942.
Kagan, B. M.: Am. J. M. Sc. **206**:309, 1943.
Kekwick, R. A.: Biochem. J. **34**:1248, 1940.
Koletsky, S., and Stecher, R. M.: Arch. Path. **27**: 267, 1939.
Kracke, R. R., and Hoffman, B. J.: Ann. Int. Med. **19**:673, 1943.
Ku, D. Y., and Simon, M. A.: Arch. Path. **18**:245, 1934.
Letterer, E.: Beitr. z. path. Anat. u. z. allg. Path. **75**: 486, 1926; Verhandl. d. deutsch. path. Gesellsch. **20**:301, 1925.
Leupold, E.: Beitr. z. path. Anat. u. z. allg. Path. **64**: 347, 1917-1918.
Loeschke, H.: Beitr. z. path. Anat. u. z. allg. Path. **77**:231, 1927.
Mahle, A. E.; Seed, L., and Welker, W. H.: Arch. Path. **26**:441, 1938.
Melnick, D.; Field, H., and Parnall, C. G.: Arch. Int. Med. **66**:295, 1940.
Morgenstern, Z.: Virchows Arch. f. path. Anat. **259**: 698, 1926.
Müller, E.: Beitr. z. path. Anat. u. z. allg. Path. **97**:42, 1936.
Murata, M., and Yoshikawa, S.: Virchows Arch. f. path. Anat. **264**:587, 1927.
Parkins, W. M.; Koop, C. E.; Riegel, C.; Vars, H. M., and Lockwood, J. S.: Ann. Surg. **118**:193, 1943.
Pauling, L.: J. Am. Chem. Soc. **62**:2643, 1940.
Pearson, B.; Rice, M., and Dickens, K.: Arch. Path. **32**:1, 1941.
Perla, D., and Gross, H.: Am. J. Path. **11**:93, 1935.
Perlzweig, W. A.; Delrue, G., and Geschickter, C.: J. A. M. A. **90**:755, 1929.
Peters, J. T.: Arch. Path. **35**:832, 1943.
Randall, O. S.: Am. J. Cancer **19**:838, 1933.
Rawson, R. A.: Am. J. Physiol. **138**:706, 1943.
Reimann, H. A.; Koucky, R. F., and Eklund, C. M.: Am. J. Path. **11**:977, 1935.
Rosenblatt, M. B.: Ann. Int. Med. **8**:678, 1934.
Rosenblum, A. H., and Kirshbaum, J. D.: J. A. M. A. **106**:988, 1936.
Rosenheim, M. L., and Wright, G. P.: J. Path. & Bact. **37**:332, 1933.
Schonholzer, G.: Klin. Wchnschr. **19**:790, 1940.
Shapiro, S.; Ross, V., and Moore, D. H.: J. Clin. Investigation **22**:137, 1943.
Shirer, J. W.; Duncan, W., and Haden, R. L.: Arch. Int. Med. **50**:829, 1932.
Strauss, A.: Virchows Arch. f. path. Anat. **291**:219, 1933.
Sweigert, C. F.: Am. J. M. Sc. **190**:246, 1935.
Ulrich, H.: Arch. Int. Med. **64**:994, 1939.
Vignati, J., and Rauchenberg, M.: Klin. Wchnschr. **16**:62, 1937.
Volland, W.: Virchows Arch. f. path. Anat. **298**:660, 1937.
White, J., and Mider, G. B.: J. Nat. Cancer Inst. **2**:95, 1941.
——Mider, G. B., and Heston, W. E.: ibid. **3**:453, 1943.
Willer, H.: Frankfurt. Ztschr. f. Path. **46**:306, 1933.
Wintrobe, M. M., and Buell, M. V.: Bull. Johns Hopkins Hosp. **52**:156, 1933.

Inflammatory Vascular Reactions.—(*a*) Allergy: During the preceding discussions the occurrence of inflammatory, fibrinoid, necrotizing lesions affecting mainly the small and the medium-sized arteries has been repeatedly mentioned in connection with, and as complications of, vasotonic as well as hydrostatic arteriosclerotic changes. It was pointed out on these occasions that apparently superimposed allergic conditions are responsible for this type of reaction. In view of the fact that these manifestations are accompanied by an infiltration of the vascular wall by plasmatic material, giving rise to fibrinoid deposits, it appears likely that the inflammatory and necrotizing variety of vascular change occurs in association with plasmatic colloidal proteinic disturbances of allergic-hyperergic nature when the immune bodies formed during these processes possess toxic properties. The hyperproteinemia occurring during immune conditions is characterized by an increase in fibrinogen and globulin (Schittenhelm) and is associated with colloidoclastic leukopenia, thrombopenia, eosinophilia, a lengthened clotting time and an accelerated rate of sedimentation of the erythrocytes during acute crises of the immune state (allergic or anaphylactic shock). Such reactions are apparently related to the degree of dispersion of the interreacting colloidal particles, as colloids consisting of highly dispersed small particles are less reactive than those with coarse particles (Klopstock).

Numerous observations connect allergic reactions with the development of degenerative arterial disease. Claims to this effect have been advanced repeatedly in regard to various vasospastic conditions (thromboangiitis obliterans, angina pectoris, reactions suggesting Raynaud's disease, malignant hypertension) caused by endogenous or exogenous physical and chemical agents, such as foodstuffs, nicotine, pollen, drugs and cold (Vaughan; Davison, Thoroughman and Bowcock; van Creveld; Werley; Sullivan and Vaughan; Krauspe; von Eiselsberg; Gaensslen; Lichtwitz; Schmidt; Conti; Dattner; Kämmerer). Joyner and Sabin reported that in certain allergic conditions the permeability of the capillary endothelium is reduced, preventing the removal of colloidal nontoxic dyes, while Klinge pointed out that anaphylactic shock can be prevented by the intravenous injection of colloidal

dyes as long as these remain in the colloidal state but not after they have precipitated in granular form. These observations on the involvement of colloidal phenomena in the production of allergic reactions are significant in view of the fibrinoid deposits found in the arterial subendothelial space in hyperergic reactions, where they may give rise to the formation of parietal thrombi, a complication especially important in the coronary arteries (Stenn; Horn and Finkelstein; Schlossmann).

The circle of the recognized or alleged allergic arterial diseases in recent years is rapidly widening, not only in the number of reported cases but also in the number of causative agents. Allergic arteritic changes occurring in connection with chronic infectious diseases, such as tuberculosis, streptococcic infections (Swift, Derick and Hitchcock; Siegmund; Strang and Semsroth; Semsroth and Koch; Karsner) and particularly rheumatic fever (Klinge; Junghanns; Masugi; Vaubel; Klinge and Vaubel; Abrikosoff; Grieshammer; Chiari; Coombs; Metz; Lieber; Von Glahn and Pappenheimer; Beneke; Abrikosoff and Rudik; Wätjen; Klotz; Geipel; Schulz and Klinge; Gross, Kugel and Epstein; Karsner and Bayless; von Sántha; Fossel; Pappenheimer and Von Glahn; Kugel and Epstein; McClenahan and Paul; Siegmund; Perla and Deutsch; Gray and Aitken; Gegenbach) have figured prominently in this respect. Serum reactions and hypersensitivity responses to therapeutically administered sulfonamide compounds have entered more recently into this field (Clark and Kaplan; Rich; Longcope), while a third group is composed of diseases with causes evidently various or unknown but suspected to be of allergic nature, such as periarteritis nodosa, thromboangiitis obliterans, isolated pulmonary sclerosis and temporal arteritis (Rössle; Jäger). There are thus hyperergic vascular reactions conditioned by general sensitizing processes and those of local character, which, however, may be of general sensitizing nature but which are locally elicited by superimposed inflammatory processes (Abrikosoff).

The hyperergic reactions of the coronary and myocardial arteries and arterioles are associated with marked endothelial proliferations encroaching on the lumens, fibrinoid masses in the subendothelial spaces, considerable edema and displacement of the muscle by connective tissue in the media, histiocytic, lymphocytic and giant cell infiltration of the interstitial tissue and perivascular collars of histiocytes, plasma cells and lymphocytes (Junghanns; Klinge; Knepper and Waaler; Vaubel; Masugi). Perivascular granulomas of the coronary vessels are found most frequently in the papillary muscle of the wall of the left ventricle and in the annulus fibrosus (Junghanns). Similar changes are encountered in the walls of the aorta, the pulmonary artery and its branches, and the renal, hepatic, cerebral and other large and small arteries (von Sántha; Stenn). Thrombi with subsequent organization and hemorrhages are seen in more acute conditions. The end result of such lesions is hyaline sclerosis or vascular obliteration by endarteritis. The chronic lesions in the stage of healing in which the inflammatory processes have subsided resemble closely those seen in other cicatricial types of arteriosclerotic changes (Chiari; Hannot; Schmitt; Jäger).

The hyperergic nature of the arterial reactions, resembling periarteritis nodosa, which were seen by Rich in 4 patients who had been treated with sulfonamide compounds, with or without injections of horse or rabbit immune serum, is supported by observations recently made by French and Weller, who found in the hearts of 126 patients who came to autopsy after treatment with sulfonamide compounds eosinophilic leukocytic infiltrations of the myocardium together with myocarditis. Similar myocardial reactions could be elicited by French and Weller in mice given intraperitoneal injections of azosulfamide, sulfanilamide, sodium sulfapyridine and sodium sulfathiazole. Rich and Gregory reproduced in rabbits arteritic lesions of the periarteritis nodosa type involving the heart, liver, kidneys, adrenal glands testes, lungs, spleen and pancreas by injecting horse serum and sulfadiazine. Similar changes were observed by Clark and Kaplan in 2 patients who died from serum sickness after an injection of horse serum. Arteritic lesions of the same type (perivascular granuloma, fibrinoid swelling of the wall, necrosis, cellular infiltration, endothelial proliferation) were produced in animals by injections of foreign protein by numerous investigators (Rich and Gregory; Vaubel; Apitz; Junghanns; Takeda; Masugi, Sato and Todo; Ceroli; Heinlein; Heinlein and Muschalik; Rintelen; Migounov; Knepper and Waaler; Fox and Jones; Stecher). Variations in extent and degree in the reactions observed by the different investigators may be related to the fact that serums of different species were used in different animals. Longcope called attention to the fact that normal and immune serums differ in antigenic properties depending on species. Practically identical inflammatory necrotizing arterial reactions were elicited in animals by repeated injections of bacterial cultures or vaccines,

especially of streptococci (Metz; Masugi and Isibasi).

(*b*) Periarteritis Nodosa: The incidence of periarteritis nodosa has undergone in recent decades considerable changes. While the majority of cases reported during the past century were reported individually, larger series of cases have been placed on record in the last four decades (von Baló). Von Baló and Nachtnebel noted in 1929 that the majority of cases on record were reported from Germany (Gruber; Jäger; Mönckeberg; Gohrbrandt; Wohlwill; Brenner; Kimmelstiel), with small numbers of cases being published in England, the United States, Australia and a few of the smaller countries. While Germany still seems to occupy the leading position, an astonishingly large number of cases have been reported from this country during recent years, of which only a few are listed here (Lund; Fitz, Parks and Branch; Malamud and Foster; Berger and Weitz; Davidsohn; Banowitch, Polayes and Charet; Allen; Coe, Reisman and DeHoff; Blaisdell and Porter; McCall. and Pennock; Motley; Felsen; Lebowich and Hunt; Jones; Weit; Wever and Perry; Haining and Kimball; Harris, Lynch and O'Hare). Jones collected in 1939 a total of 101 cases recorded in the English literature.

The disease affects persons of all ages and small and large vessels, irrespective of the presence of vasa vasorum (Lange). Of a total of about 230 patients whose cases were reported up to 1937, 32 (13.9 per cent) were children (Coe, Reisman and DeHoff). The mesenteric (Haining and Kimball) and renal vessels were apparently most often affected (80 per cent of the cases, Arkin; 100 per cent, McCall and Pennock), and nephrosclerosis, therefore, often accompanied the disease. Periarteritis nodosa may affect the vessels of individual organs only —for example, pulmonary vessels (Sternberg; Rössle) or renal vessels (Hauser)—or may involve practically the entire arterial and sometimes also the venous system. In view of the frequently acute character of the arteritic processes multiple aneurysms are often present. The disease, which often appears after acute infections, usually takes a relatively rapid and fatal course, but some patients survive for several years, and occasional ones are cured. The disease occurs also in animals—cattle (Hoogland; Guldner; Nieberle), hogs (Joest; Joest and Harzer; Hoogland; Henschen; Nieberle), deer (Lüpke; Jäger) and dogs (von Baló). Eosinophilia of the blood is often observed and is sometimes marked.

Attempts aimed at the isolation of specific causative agents from the tissues and the blood of affected persons have failed (Gohrbrandt; von Hann). The claim made by von Hann as to the experimental reproduction of the disease in guinea pigs given injections of blood from a patient is based on a misinterpretation of normal conditions in the lungs of guinea pigs (Lemke). Selye and Pentz recently advanced the theory that periarteritis nodosa, rheumatic arteritis and nephrosclerosis may be causally related in part to abnormal and probably excessive adaptive responses of the adrenal cortex.

(*c*) Temporal Arteritis: It is likely that the giant cell chronic arteritis of the temporal artery belongs to this complex of allergic arterial reactions. Chasnoff and Vorzimer considered it a local manifestation of a systemic disease usually ending after a number of months in recovery but occasionally causing death. The lesions in the temporal arteries differ histologically sufficiently from those seen in periarteritis nodosa and rheumatic arteritis to make them a distinct entity. The changes, however, are not limited to the temporal arteries but occur also in the carotid, central retinal, cerebral, occipital and radial arteries. Similar giant-cellular necrotizing granulomatous medial changes affecting the aorta, the branches of the aorta and their branches have been seen by Gilmour in 4 cases. Sproul and Hawthorne reported similar lesions in the aorta and the iliac arteries of 2 patients. Hoyt, Perera and Kauvar and also Weiss recorded cases of temporal arteritis and the changes considered as common local manifestations of a generalized arterial disease. Additional reports dealing with temporal arteritis have been made (Horton, Magath and Brown; MacDonald and Moser; Bowers; Dick and Freeman; Jennings; Bain; Horton and Magath; Sprague and Mackenzie), making a total of 21 cases. The disease usually involves elderly persons, more often those of the female sex. The lumens of the rigid vessels are narrowed or obliterated by proliferation of the intima. The media is invaded by monocytes and replaced in parts by a granulomatous tissue containing giant cells. Thrombi occur occasionally. Aneurysmal sacs may arise in the media.

COLLOID PLASMATIC DISTURBANCES: C. PROTEINIC TYPE

Inflammatory Vascular Reactions:

Abrikosoff, A. I.: Virchows Arch. f. path. Anat. **295**: 669, 1935.

——and Rudik, D.: Arch. internat. de méd. expér. **10**: 303, 1935.

Allen, P. D.: Arch. Surg. **40**:271, 1940.

Apitz, K.: Virchows Arch. f. path. Anat. **289**:46, 1933.

Bain, C. W. C.: Lancet **1**:517, 1938.

von Baló, J.: Virchows Arch. f. path. Anat. **248**:337, 1924.

——and Nachtnebel, E.: ibid. **272**:478, 1929.
Banowitch, M. M.; Polayes, S. H., and Charet, R.: Ann. Int. Med. **16**:1149, 1942.
Beneke, R.: Virchows Arch. f. path. Anat. **254**:723, 1924.
Berger, S. S., and Weitz, M. A.: J. Allergy **9**:489, 1938.
Blaisdell, E. R., and Porter, J. E.: New England J. Med. **224**:1087, 1941.
Bowers, J. M.: Arch. Int. Med. **66**:384, 1940.
Brenner, F.: Frankfurt. Ztschr. f. Path. **51**:479, 1938.
Chasnoff, J., and Vorzimer, J. J.: Ann. Int. Med. **20**: 327, 1944.
Chiari, H.: Beitr. z. path. Anat. u. z. allg. Path. **80**: 336, 1928; **88**:1, 1931; Klin. Wchnschr. **9**:1862, 1930.
Clark, E., and Kaplan, B. I.: Arch. Path. **24**:458, 1937.
Coe, M.; Reisman, H. A., and DeHoff, J.: J. Pediat. **18**:793, 1941.
Conti: Atti Soc. lomb. sc. med. e biol. **18**:121, 1929.
Coombs, C.: Quart. J. Med. **2**:26, 1908.
van Creveld, S.: Ann. pædiat. **157**:84, 1941.
Dattner, B.: Nervenarzt **4**:573, 1931.
Davidsohn, I.: Illinois M. J. **83**:352, 1943.
Davison, H. M.; Thoroughman, J. C., and Bowcock, H.: South. M. J. **36**:560, 1943.
Dick, G. F., and Freeman, G.: J. A. M. A. **114**:645, 1940.
von Eiselsberg, K. P.: Klin. Wchnschr. **12**:1174, 1933; **13**:619, 1934; Wien. klin. Wchnschr. **45**:332, 1932.
Felsen, J.: Ann. Int. Med. **15**:251, 1941.
Fitz, R.; Parks, H., and Branch, C. F.: Arch. Int. Med. **64**:1133, 1939.
Fossel, M.: Frankfurt. Ztschr. f. Path. **54**:588, 1940.
Fox, R. A., and Jones, L. R.: Proc. Soc. Exper. Biol. & Med. **55**:294, 1944.
French, A. J., and Weller, C. V.: Am. J. Path. **18**: 109, 1941.
Gegenbach, A.: Zentralbl. f. allg. Path. u. path. Anat. **36**:244, 1925.
Geipel, P.: München. med. Wchnschr. **54**:1057, 1907; **56**:2469, 1909; Deutsches Arch. f. klin. Med. **85**:75, 1905.
Gilmour, J. R.: J. Path. & Bact. **53**:263, 1941.
Gohrbrandt, P.: Virchows Arch. f. path. Anat. **263**: 246, 1927.
Gray, S. H., and Aitken, L.: Arch. Path. **8**:451, 1929.
Grieshammer, W.: Klin. Wchnschr. **18**:451, 1939; Frankfurt. Ztschr. f. Path. **53**:136, 1939.
Gross, L.; Kugel, M. A., and Epstein, E. Z.: Am. J. Path. **11**:253, 1935.
Gruber, G. B.: Virchows Arch. f. path. Anat. **245**:123, 1923; **248**:441, 1925; Verhandl. d. deutsch. path. Gesellsch. **19**:313, 1923.
Haining, R. B., and Kimball, T. S.: Am. J. Path. **10**:349, 1934.
von Hann, F.: Virchows Arch. f. path. Anat. **227**:90, 1919.
Harris, A. W.; Lynch, G. W., and O'Hare, J. P.: Arch. Int. Med. **63**:1163, 1939.
Heinlein, H.: Arch. f. exper. Path. u. Pharmakol. **179**: 127, 1935; Virchows Arch. f. path. Anat. **290**:667, 1937.
——and Muschalik: Klin. Wchnschr. **16**:873, 1937.
Horton, B. T., and Magath, T. B.: Proc. Staff Meet., Mayo Clin. **12**:548, 1937.
——Magath, T. B., and Brown, G. E.: Arch. Int. Med. **53**:400, 1934.
Hoyt, L. H.; Perera, G. A., and Kauvar, A. J.: New England J. Med. **225**:283, 1941.
Jaeger, A.: Verhandl. d. deutsch. path. Gesellsch. **13**: 209, 1909; Virchows Arch. f. path. Anat. **197**:71, 1909.
Jäger, E.: Virchows Arch. f. path. Anat. **284**:526, 1932; **288**:833, 1933.
Jennings, G. H.: Lancet **1**:424, 1938.
Joest, E., and Harzer, J.: Beitr. z. path. Anat. u. z. allg. Path. **69**:85, 1921.
Jones, G. M.: Ann. Int. Med. **16**:920, 1942.
Joyner, A. C., and Sabin, F. R.: J. Exper. Med. **68**: 325, 1938.
Junghanns, E.: Beitr. z. path. Anat. u. z. allg. Path. **92**:467, 1933.
Kämmerer, H.: Allergische Diathese und allergische Erkrankungen, Munich, J. F. Bergmann, 1934, p. 271.
Karsner, H. T.: Am. J. Path. **10**:704, 1934.
——and Bayless, F.: Am. Heart J. **9**:551, 1934.
Kimmelstiel, P.: Virchows Arch. f. path. Anat. **265**: 16, 1925.
Klinge, F.: Klin. Wchnschr. **6**:2265, 1927; **9**:586, 1930; Beitr. z. path. Anat. u. z. allg. Path. **83**:185, 1930; Virchows Arch. f. path. Anat. **278**:438, 1930; **279**:1, 1930.
——and Vaubel, E.: ibid. **281**:701, 1931.
Klinger, H.: Frankfurt. Ztschr. f. Path. **42**:455, 1931.
Klopstock, A.: Immunität, in Lichtwitz, L.; Liesegang, R. E., and Spiro, K.: Medizinische Kolloidlehre, Leipzig, Theodor Steinkopff, 1935, p. 193.
Knepper, R., and Waaler, G.: Virchows Arch. f. path. Anat. **294**:587, 1935.
Krauspe: Klin. Wchnschr. **15**:1742, 1936.
Kugel, M. A., and Epstein, E. Z.: Arch. Path. **6**:247, 1928.
Lange, F.: Virchows Arch. f. path. Anat. **248**:463, 1924.
Lebowich, J., and Hunt, H. D.: Am. J. Clin. Path. **10**:542, 1940.
Lemke, R.: Virchows Arch. f. path. Anat. **245**:322, 1923.
Lichtwitz: Klin. Wchnschr. **4**:2353, 1925.
Lieber, M. M.: Beitr. z. path. Anat. u. z. allg. Path. **91**:594, 1933.
Longcope, W. T.: Medicine **22**:251, 1943.
Lund, H. Z.: Ohio State M. J. **38**:244, 1942.
McCall, M., and Pennock, J. W.: Am. J. M. Sc. **206**: 652, 1943.
McClenahan, W. U., and Paul, J. R.: Arch. Path. **8**:595, 1929.
MacDonald, J. A., and Moser, R. H.: Ann. Int. Med. **10**:1721, 1937.
Malamud, N., and Foster, D. B.: Arch. Neurol. & Psychiat. **47**:828, 1942.
Masugi, M.: Beitr. z. path. Anat. u. z. allg. Path. **91**: 82, 1933; Klin. Wchnschr. **14**:373, 1935.
——and Isibasi, T.: Beitr. z. path. Anat. u. z. allg. Path. **96**:391, 1936.
——and Yä-Shu: Virchows Arch. f. path. Anat. **302**: 39, 1938.
——Sato, Y., and Todo, S.: Tr. Soc. path. jap. **25**: 211, 1935.
Metz, W.: Beitr. z. path. Anat. u. z. allg. Path. **88**:17, 1931.
Migounov, B. I.: Acta rheumat. **6**:9, 1934.
Mönckeberg, J. G.: Beitr. z. path. Anat. u. z. allg. Path. **38**:101, 1905.
Motley, L.: J. A. M. A. **106**:898, 1936.
Nieberle, K.: Virchows Arch. f. path. Anat. **256**:131, 1925; **269**:587, 1928.
Pappenheimer, A. M., and Von Glahn, W. C.: Am. J. Path. **3**:583, 1927; **2**:15, 1926; J. M. Research **44**: 489, 1924.

Perla, D., and Deutsch, M.: Am. J. Path. **5**:45, 1929.
Rich, A. R.: Bull. Johns Hopkins Hosp. **71**:123, 1942. ——and Gregory, J. E.: ibid. **72**:65, 1943.
Rintelen, W.: Virchows Arch. f. path. Anat. **299**:629, 1937.
Rössle, R.: Virchows Arch. f. path. Anat. **288**:780, 1933; Klin. Wchnschr. **12**:574, 1933.
von Sántha, K.: Virchows Arch. f. path. Anat. **287**: 405, 1932.
Schittenhelm, A., in Mohr, L., and Staehelin, R. Handbuch der inneren Medizin, ed. 2, Berlin, Julius Springer, 1925, vol. 1, p. 1.
Schlossmann, N. C.: Arch. Path. **34**:365, 1942.
Schmidt, R.: München. med. Wchnschr. **77**:1435, 1930.
Schmitt, H.: Virchows Arch. f. path. Anat. **296**:603, 1936.
Schulz, M., and Klinge, F.: Virchows Arch. f. path. Anat. **288**:717, 1933.
Selye, H., and Pentz, E. I.: Canad. M. A. J. **49**:264, 1943.
Semsroth, K., and Koch, R.: Krankheitsforsch. **8**:191, 1930.
Siegmund, H.: Zentralbl. f. allg. Path. u. path. Anat. **32**:276, 1924; **51**:385, 1931; Verhandl. d. deutsch. path. Gesellsch. **19**:114, 1923; Ztschr. f. Kreislaufforsch. **21**:389, 1929.
Sprague, P. H., and Mackenzie, W. C.: Canad. M. A. J. **43**:562, 1940.
Sproul, E. E., and Hawthorne, J. J.: Am. J. Path. **13**:311, 1937.
Stecher, W.: Virchows Arch. f. path. Anat. **300**:645, 1937.
Stenn, F.: Arch. Path. **26**:244, 1938.
Strang, J. M., and Semsroth, K.: Arch. Int. Med. **47**: 583, 1931.
Sullivan, C. I., and Vaughan, W. T.: J. Allergy **9**:48, 1937.
Swift, H. F.; Derick, C. L., and Hitchcock, C. H.: J. A. M. A. **90**:906, 1928.
Takeda, K.: Tr. Soc. path. jap. **25**:194, 1935.
Vaubel, E.: Beitr. z. path. Anat. u. z. allg. Path. **89**: 374, 1932; **83**:185, 1929.
Vaughan, W. T.: The Practice of Allergy, St. Louis, C. V. Mosby Company, 1939, p. 1014.
Von Glahn, W. C., and Pappenheimer, A. M.: Am. J. Path. **2**:235, 1926.
Wätjen: Verhandl. d. deutsch. path. Gesellsch. **18**:223, 1921.
Weiss, S.: New England J. Med. **225**:579, 1941.
Werley, G.: J. Allergy **4**:65, 1932; Southwest. Med. **22**:431, 1938.
Wever, G. K., and Perry, I. H.: J. A. M. A. **104**: 1390, 1935.
Wevi, D. R.: Am. J. Path. **15**:79, 1939.
Wohlwill, F.: Virchows Arch. f. path. Anat. **246**:377, 1923.

HEMATIC ANOXEMIA

Agents that cause the production of inert hemoglobin derivatives, such as methemoglobin, carbon monoxide–hemoglobin and sulfmethemoglobin, which lack the oxygen-carrying power of normal hemoglobin, interfere with the proper oxygenation of the vascular wall and may thereby give rise to the development of degenerative changes in the vascular wall. Inasmuch as many of these agents exert also a vasotonic effect, their arteriosclerotogenic action is based on the summation of two different causative mechanisms. Agents of this type are carbon monoxide, nitrites and sulfonamide compounds. It is likely that the medial necrosis in the aortas of rabbits produced by Loewe, Jürgens and Noltemeier by the intravenous injection of chloramine and by Mancke and Droop by the oral administration of formaldehyde or formaldehyde sodium bisulfite is in part a result of the formation of methemoglobin. A similar mechanism was probably active in the production of medial necroses in the arteries of rabbits following the intravenous injection of chloropicrin and benzoyl peroxide (Müller) and of hydrogen peroxide, magnesium peroxide, benzoyl peroxide, acetylchloroammonium benzol, hydroquinone, quinone and methylthionine chloride (Rieder; Siebert), and after the percutaneous application of toluene sulfodichloroamide, dichloroamidobenzoic acid, acetylchloroaminobenzol and chloroamido carbonic ethyl ester.

A similar hematic effect on the oxygenation of the vascular walls occurs if the blood is not completely saturated with oxygen while passing through the lungs because of reduced atmospheric oxygen pressure. Campbell, as mentioned before, reported that animals kept for several weeks in an atmosphere with greatly reduced oxygen tension showed intimal thickening of their pulmonary arterioles. However, a greatly increased oxygen pressure of the atmospheric air also interferes with adequate oxygenation of the tissues by causing an imbalance between the carbon dioxide and the oxygen tension in the tissues and an accumulation of carbon dioxide in the tissues and by eliciting thereby hyperoxemic hypoxidosis (Strughold; Gsell; Bean and Bohr; Hinshaw and Boothby). This mechanism is in part responsible for the intimal thickenings of the pulmonary arteries observed in rats subjected for more than twenty-four days to inhalation of compressed air with an oxygen tension of 635 mm. of mercury (Bennett), and such disturbances in the carbon dioxide–oxygen balance of the blood and tissues are associated also with vasculotonic reactions which accentuate the hematic anoxemic arteriosclerotogenic effects. These anoxemic reactions are, moreover, responsible for the development of myocardial degenerations in oxygen poisoning (Kaunitz).

HEMATIC ANOXEMIA

Droop, H.: Experimental Untersuchungen an Kaninchen über die Stoffwechselgenese der Arteriosklerose und die diätetische Beeinflussung dieses Prozesses, Inaug. Dissert., Göttingen, W. F. Kaestner, 1915.
Gsell, R.: Am. J. Physiol. **66**:5, 1923.

Hinshaw, H. C., and Boothby, W. M.: Proc. Staff Meet., Mayo Clin. **16**:211, 1941.
Kaunitz, J.: J. Aviation Med. **13**:267, 1942.
Loeb, O.: Arch. f. exper. Path. u. Pharmakol. **69**:114, 1912.
Mancke, R.: Arch. f. exper. Path. u. Pharmakol. **141**: 228, 1929.
Müller, W.: Weitere Beiträge zur Kenntnis der resorptiven Wirkung der Oxydationsmittel, Inaug. Dissert., Göttingen, E. Hofer, 1919.
Noltemeier, H. H. T.: Beiträge zur experimentellen Pharmakologie des disponiblen chlors, Inaug. Dissert., Göttingen, E. Hofer, 1919.
Rieder: Ztschr. f. d. ges. exper. Med. **10**:169, 1920.
Rossmanns: Inaug. Dissert., Göttingen, 1920.
Siebert: Ztschr. f. d. ges. exper. Med. **9**:123, 1919.
Wenzel, H.: Arch. f. exper. Path. u. Pharmakol. **137**: 215, 1928.

PREVENTIVE AND THERAPEUTIC ASPECTS

In his recent treatise on vascular scleroses Moschcowitz came to the conclusion that

> . . . unless the individual is cut down by intercurrent disease, death is invariably the result of arteriosclerosis. It is the inevitable destiny of all creatures who possess a cardiovascular system with intravascular pressure. . . . Arteriosclerosis being an inevitable consequent of ageing and therefore an irreversible process, it is hardly likely that any method of therapy will ever be discovered which will restore the diseased vessels to their normal texture, unless we can cure mortality. Nor can arteriosclerosis be prevented, no more than gray hair or facial wrinkles.

This fatalistic attitude based on the unrestricted acceptance of the old age theory of arteriosclerosis finds its equivalent in, and has as much merit as, the one propounded by the proponents of the old age theory of cancer, who are equally emphatic in their dictum that cancer is the ultimate cause of death of all persons living sufficiently long. It was pointed out recently by Hueper that there exists a sufficient amount of reliable and valid evidence showing that cancer is not an obligatory outcome of senescent tissue changes but is a reaction to abnormal exogenous and endogenous factors of physical and chemical nature. It must be equally clear from the experimental and clinical data presented here that also arteriosclerosis in its various types is essentially not an old age disease but a condition brought about by various exogenous and endogenous agents and influences acting on the blood and the vessels during the major part of life and independent of any senescence process. Cancer and arteriosclerosis are definite disease phenomena and not physiologic or pathologic manifestations of old age. They are for this reason amenable to preventive and therapeutic measures. The fact that the means at present at one's disposal are defective and in part irrational does not detract from the validity of the foregoing statement but should provide inspiration for the development of improved measures.

It is felt that the preceding presentation dealing with the numerous causes and the different causative mechanisms and their related anatomic vascular manifestations furnishes an adequate basis for the development of rational diagnostic, preventive and therapeutic methods. The evidence presented, however, definitely indicates that there is no single approach to this problem which will be equally effective for the preventive and the therapeutic management and the diagnosis of the different types of degenerative and sclerosing vascular lesions.

The diagnostic methods and the therapeutic or preventive measures needed for the demonstration and the treatment of the hematic changes underlying and preceding the development of the vascular reactions caused by colloidal plasmatic disturbances will be different from those indicated for the discovery and the therapy of the functional and the anatomic vascular and organic reactions associated with arteriosclerosis on the basis of vasotonic and hydrostatic factors. In planning the control of these vascular diseases consideration must be given to the facts that arteriosclerosis is a disease complex that starts its anatomic development during middle adult life and that all therapeutic measures are utterly incapable of restoring an arterial system with cicatricial arteriosclerotic lesions in which the functioning elastic and muscular elements of the vascular wall are replaced by nonfunctioning fibrous, hyaline and calcified matter to its original state of functional and anatomic integrity. Only in the early stages of the disease can it be cured, while in advanced stages merely its further progress can be arrested or impeded (Plesch). It is pertinent, therefore, that any preventive measures must be started at a time when there are only functional vascular or hematic changes present, i. e., during the early part of adult life. It should be equally clear that effective diagnostic supervision and preventive management of the patient should from then on extend over the remaining part of life if sickness, disability and death from arteriosclerotic disease is to be avoided or to be appreciably limited.

While it is beyond the scope of this presentation to deal in detail with the prevention and the therapy of arteriosclerosis it seems advisable to indicate the general lines of approach by which this goal might be achieved as they appear from this investigation. Measures directed at the prevention of the deposition of lipoid material in the arteries must aim at keeping the plasma cholesterol level within the lower part of the normal

range or, if that is not possible because of some uncontrollable factor, at improving the stability of the colloidally dispersed cholesterol so as to prevent its precipitation in and on the vascular walls. The reduction of the cholesterol level of the plasma may be attained by limiting the alimentary intake of cholesterol-containing foodstuffs (Tuohy; Leary) and by administering blood cholesterol–lowering agents, such as, especially, thyroid preparations and, in diabetic conditions, possibly, lipocaic in conjunction with insulin. The stabilization of the cholesterol sol in the plasma at a safe level may be accomplished by the oral introduction or the parenteral injection of peptizing agents which increase the colloidal dispersion of the cholesterol and thereby lower its precipitability. It is possible that the favorable effect seen from the use of thiocyanides and iodine compounds of fatty acids (diiodine ricinstearolate) in experimental cholesterol atheromatosis depends on this mechanism. Consideration should be given in this respect also to the lecithin and bile acid content of the plasma. The more difficult problem of a rapid and effective mobilization of lipoid material in the atheromas and its removal with the blood, which is especially important in atheromatosis of the coronary arteries, may perhaps be approached by applying in vivo the methods used in ordinary life for the removal of greasy material from fabrics through the employment of colloidal emulsifiers and detergent solubilizers. However, it should be emphasized in this connection that a great deal more information on the factors controlling the colloidal equilibrium of the plasma is necessary before real progress in this field will be likely.

Recent experiences with the experimental production of vascular diseases by changes in the plasma proteins direct attention to certain so far neglected aspects of modern chemotherapy with sulfonamide compounds and related compounds. It seems to be necessary that more attention be paid to the possibility of allergic vascular complications arising from such procedures. The observations made on such occasions suggest, moreover, that vitaminic imbalances resulting from such therapy and apparently affecting mainly the vitamin B complex require closer study as to their arteriosclerotogenic potentialities.

The occurrence of degenerative and cystic medial lesions in the aorta as late results of severe hypotonic episodes make it urgent that conditions of circulatory failure, such as those seen in shock, are cut as short as possible with the existing therapeutic measures. Persons who have passed through prolonged and marked hypotensive states should be cautioned against any excessive strain to their vascular system, particularly during the early period following such an attack, i. e., before firm scar tissue may have formed.

The prevention of chronic arterial lesions on the basis of occupational or environmental exposure to vasotonic chemicals with hypotonic (carbon monoxide, nitrites, cyanides) or hypertonic effect (digitalis, viosterol, ephedrine and derivatives, nicotine, lead) is mainly a matter of industrial and public hygiene and thus a subject of legislation and education.

It is obvious that adequate preventive measures cannot always be instituted in time, because the exposure to the etiologic vasotonic agents is not properly recognized or cannot be combated for various technical reasons or is disregarded out of negligence. The development of the ultimately appearing degenerative and sclerosing arterial lesions raises then the question of the use of appropriate therapeutic measures. The type of therapeutic measure adopted depends on the character of the causative arteriosclerotogenic vasotonic mechanism present and on the kind of therapeutic action desired or possible in the individual case. Three different types of therapeutically active agents may be chosen for this purpose if available:

1. Agents which destroy the vasotoxic principle by oxidation, reduction, hydrolysis, conjugation or other types of metabolic disintegration or alteration or which make the vasotoxic principle innocuous and ineffective by blocking its site of action (structural blocking).

2. Agents which eliminate or increase the excretion of a causative vasotonic principle. The administration of calcium salts, viosterol and parathyroid hormone in chronic lead poisoning or the removal of parathyroid adenomas in hyperparathyroidism belong in this category.

3. Agents which exert a vasotonic effect opposite to that exerted by the causative principle. This type of therapeutic measure is obviously the least desirable and effective one, as it usually merely produces symptomatic relief without influencing in any way the continued existence of the causal conditions. Vasodilating agents, such as nitrites, thiocyanides, theobromine, acetylcholine and similarly acting substances, are used in the symptomatic control of hypertension of various genesis, while epinephrine, ephedrine, extract of adrenal cortex and other hypertonic agents are employed in counteracting hypotensive states of diverse nature. Inasmuch as the site of action of a vasculotonic agent depends on its type and on

the dose administered, it is advisable to select for therapeutic purposes a vasotonic agent which exerts its effect in the same region in which the causal arteriosclerotogenic vasotonic agent is operative. Disregard of this principle may result in apparent symptomatic relief through the production of abnormal functional and, finally, anatomic changes in some other part of the cardiovascular system. To lower increased blood pressure caused by vascular hypertonia through interference with cardiac function resulting in a reduction of the cardiac output is thus not only an irrational but a harmful procedure.

It is evident from these considerations that the available therapeutic measures are of only limited value, as at best they do no more than prevent any further progress of the arteriosclerotic process. In general they restrict to some extent the unhampered advance of the vascular changes or simply produce some temporary symptomatic relief without interfering seriously with the development of these changes.

REFERENCES FOR PREVENTIVE AND THERAPEUTIC ASPECTS

Hueper, W. C.: Occupational Tumors and Allied Diseases, Springfield, Ill., Charles C Thomas, Publisher, 1942.

Moschcowitz, E.: Vascular Scleroses, New York, Oxford University Press, 1942, p. 178.

Plesch: Klin. Wchnschr. **11**:524, 1932.

Tuohy, E. L.: J. A. M. A. **121**:42, 1942.

Book Reviews

The Biological Basis of Individuality. By Leo Loeb, professor emeritus of pathology, Washington University School of Medicine, St. Louis. Price $10.50. Pp. 711. Springfield, Ill.: Charles C Thomas, Publisher, 1944.

The comprehensiveness of this book makes it difficult to review all parts of it with equal adequacy; an attempt, therefore, is made to convey to the reader of this review only the main gist of the book in which the author has synthesized the results of his own studies and those of related investigations by other biologists into a highly original and far reaching system.

Individuality is the original physical and psychic state of an organism which has developed in accordance with its genetic constitution in cooperation with environmental factors. The concept of a biologic basis of individuality originated in the well known investigations on transplantation which the author has conducted during the past five decades. Transplantation of tissue from one animal to another calls forth a cellular reaction which is the stronger the greater the strangeness between host and donor. Only autogenous grafts fail to elicit such a reaction, but even if host and donor are as closely related as litter mates of a strain of mice inbred through many generations, the transplants will produce a mild reaction in the host. The results of the latter syngenesiotransplantations indicate incidentally that complete homozygosity has as yet not been achieved by continuous inbreeding.

There are indications that the reaction around a graft is caused by certain proteins, probably by nucleoproteins given off by the transplant: Each individual is the carrier of certain proteins or of a certain protein which distinguishes all the organs and tissues of this individual from the organs and tissues of any other individual of the same species and strain. This distinctive substance, termed "individuality differential," is present in all tissues and organs and also in the blood of an individual. Besides this individuality differential, each organism carries a number of substances typical of the strain, the species or the class to which it belongs and which distinguish it from the members of all other strains, species or classes. All these distinctive substances, including the individuality differential, are designated as "organismal differentials."

Species differentials are responsible for the marked reaction that a heterotransplant elicits in the host leading to quick destruction of the graft. The violence of this reaction makes heterotransplantation less suitable for the detection of species differences than serologic methods which demonstrate the presence of antibodies in the serum of immunized animals. For the detection of finer differences between individuals, transplantation is not only superior to serologic methods but the method of choice, although individuality differentials have been shown to exist in the red corpuscles of certain animals. In addition, there are the organ and tissue differentials. They are responsible for the difference in the reactions elicited in the host by various tissues of the same individual.

Data on the responses of the host to various types of transplants and on the relation of the differentials to varying conditions in the host (age, hormonal influences, blood groups) occupy almost one fourth of the book and include much new material. In conclusion two distinct types of biologic individuality are recognized: 1. The "mosaic individuality" based on the sum of the organ and tissue differentials carried by an individual. This is the individuality recognized so far and studied in particular by the geneticists. 2. The "essential individuality" based on the distinction between the individuality differential of one individual from the individuality differentials of all other individuals. Both mosaic and essential individuality depend primarily on genetic factors, but they are also under the influence of environmental conditions.

Subsequently, the phylogenetic and the ontogenetic development of the differentials is traced from lower organisms, such as coelenterates, planaria and amphibians to birds and mammals. In the course of evolution of animals a finer differentiation takes place and a more rigid specificity of the differentials ensues. Thus in primitive species heterotransplantation even of whole organs may succeed, and two individuals may be joined together without development of signs of incompatibility such as occurs, for instance, in parabiotically joined rats. Corresponding to the phylogenetic evolution of the differentials there is a progressive ontogenetic development of the latter. Against embryonic tissues reactions may be lacking or slight, and tissues of young individuals produce milder reactions in the host than those of adults. Thus, in young organisms the proteins representing the individuality differentials have not as yet achieved as high a specificity as in the adult. There is, therefore, an inverse relation between the adaptability of an organism and the rigidity of its individuality. A certain relation exists also between the development of the organismal differentials and the organizers: With advancing embryonal development, the organizers are replaced by complex highly specific contact substances which maintain the normal equilibrium between neighboring tissues and organs. Graded manifestations of compatibility or incompatibility between unicellular organisms suggest that even in these primitive structures there may exist finely differentiated substances analogous to the organismal differentials of higher species. However, these substances are not identical with true organismal differentials. From this point of view the author analyzes the reactions taking place between individual protozoa and those between spermatozoon and egg; he also discusses the interaction between single cells during the processes of tissue formation from the primitive amebocytes of Limulus to the cells of higher organisms.

Transplantation as a systematic method for tumor research was first used by the author in 1901. Tumors are bearers of the same or almost the same organismal and individuality differentials as the normal organs and tissues of the individual in which they originated. The better transplantability of tumors, i. e., the successful transplantation into homoiogenous and even heterogenous animals is due largely to the increased growth energy of the tumor and to processes of adaptation between host and donor. These processes are fully

discussed with particular reference to the production of immunity to transplanted tumors. The author holds the view that the development of a spontaneous tumor is not due to a somatic mutation. This interpretation agrees with the conclusion that factors underlying transplantability of tumors are not the same as those involved in spontaneous tumor growth.

The following part of the book deals with the nature of the organ and tissue differentials and with their mutual relations, and also with the relations between organ and organismal differentials. It has as yet not been possible to isolate these antigenic differentials by chemical means. They are, however, not identical with any of the known antigens which have so far been analyzed in studies on blood groups, Rh factor, precipitins, hemolysins or cytotoxins. They are genetically determined but are not present in the genes; the latter contain or may determine the formation of precursor substances which during ontogenesis develop into the differentials proper.

The presence of the same individuality differential in all organs and tissues of an individual makes possible the normal interaction between the various tissues and between tissues and body fluids. In order to interact harmoniously, the various tissues and body fluids which represent the inner environment of the body have to be specifically adapted to one another. Thus there is in the healthy organism an "autogenous equilibrium," meaning identity of the individuality differentials in tissues and body fluids of the same individual. Disharmonies in this balance give rise to disease. Similarly, aging represents a continuous decline of the mutual adaptation between the constituent parts of an organism, ultimately leading to death. The lower the species, the fewer and less complex are the tissues and substances involved in the maintenance of a normal equilibrium. In lower organisms not only entire organs but even a whole new organism may be regenerated from parts of the old one after injury. These lower organisms are thus potentially immortal. Higher organisms have developed such a complex system of structure and function that they have lost their adaptability to unfavorable environmental conditions. They have become rigid and have acquired senescence and disease; they have attained refinement of individuality but at the expense of immortality.

While thus in the course of evolution the higher organisms have lost to a large extent their power of adaptation to the outer environment, the central nervous system and higher sense organs which serve as a means of communication between the individual and his environment have developed a high specialization and extended their significance. In the sphere of the psychic aspects of the individuality, the organism has become more dependent on the environment. These considerations lead to the final chapter of the book dealing with the psychosocial aspect of individuality. The psychic attitude of an individual is in the last analysis determined by reactions of his nervous system which in turn is also a carrier of his individuality differential. This being genetically determined, all actions and emotions of an individual are likewise under the influence of his genetic constitution. However, environmental conditions in higher organisms play a prominent role in determining psychic and social attitudes. This is in contrast to the physical makeup of the individual which is preponderantly influenced by genetic factors and only to a lesser extent by environmental conditions.

As with many a masterwork on biology, objections may possibly be raised by some reviewers to one or the other of the author's interpretations. This would not in the least affect the significance and value of the book, which constitutes a unique presentation of the meaning of individuality on the basis of present knowledge in biology. A wealth of information for the general pathologist and the biologist is provided by this book, which is particularly valuable since the author deals in an unusually objective manner with the results of other investigators and their views. A discussion of the many problems of immunity and genetics dealt with in full and correlated with the author's concept would be beyond the scope of this review. The book will serve as a stimulus to further investigations by open-minded experimentalists. To the general pathologist it is another step in the further development and modification of the concepts of Virchow. The student in the fields of transplantation, and tissue and tumor growth will welcome this comprehensive and for the pathologist indispensable work.

A Textbook of Pathology. By Robert Allan Moore, Edward Mallinckrodt professor of pathology, Washington University School of Medicine, St. Louis. Pp. 1338, with 513 illustrations. Price $10. Philadelphia and London: W. B. Saunders Company, 1944.

To justify its appearance in times like these a new textbook should have great merit. In the opinion of the reviewer this book is more than justified.

The division of the subject matter into general and special pathology, usually found in textbooks of pathology, is followed in this book, but the relative amount of space devoted to each is unusual. General pathology is covered in only 207 pages, while 1,084 are devoted to special pathology. The organization of the material in both sections is new, as is the manner of presentation.

In the part on general pathology the greatest innovation is in the manner of dealing with the retrogressive and degenerative changes. Here the classification is not morphologic but chemical. Chapters are devoted to disturbances in the metabolism of proteins, carbohydrates, lipids and minerals and in the fluids of the body. The organization of the material on inflammation, tumors and disturbances in circulation is more conventional, but the emphasis is on the physiologic and chemical aspects.

The material in special pathology is divided on the basis of whether the cause is known. The diseases of known cause are discussed in parts devoted to those caused, respectively, by living agents, physical agents, chemical agents and deficiencies and those related to pregnancy and the fetal and newborn states. The diseases of unknown or obscure cause are presented in the last part of the book, where they are taken up by systems in the usual way. This is the largest part (544 pages), and together with that on diseases caused by living agents (398 pages) it constitutes most of the volume.

This organization of material has numerous advantages, but it leads to some difficulties and inconsistencies. Thus discussions of diseases of some particular organs are located in widely separated chapters; for example, the renal diseases. Diseases caused by living agents are classified in various ways: according to portal of entry, source of the infection, class of organisms or method of transmission. In general this works out well, but some diseases logically belong in several places, having more than one portal of entry, method of spread, etc., and others get misplaced. Thus, although pneumonia is the chief lesion produced by Klebsiella pneumoniae, this organism is discussed with the intes-

tinal bacteria. At other times diseases become misplaced because there are not enough chapter headings to go around. Thromboangiitis obliterans and Raynaud's disease are placed in the chapter on arteriosclerosis, to the possible confusion of the student.

This book is remarkably comprehensive and up to date. Some of the exotic infections are presented in considerable detail. The style of writing, while not always fluent, is lucid, readable and to the point. The advisability of beginning the presentation of new diseases abruptly with the pathologic anatomy might be questioned on pedagogic grounds. There are numerous references at the end of each chapter and, what is a commendable innovation, the names of pertinent journals.

This book has so many good points and it represents such a laudable effort to inject new life into the subject by a new approach that the reviewer hesitates to point out some minor deficiencies. The author of a textbook must make many decisions on the apportioning of space to the various subjects, and with his decisions others may disagree. Whether 46 pages should be devoted to syphilis and only 28 to tuberculosis and 46 to tumors may be questioned. The infectious granulomas are not presented as a group. The wisdom of abandoning this classification is questioned. In the presentation of edema, the factors in its causation are given, but there is no discussion of the general types, the hazards to the individual patient or the chronic effects in the tissues or serous cavities. In the discussion of hemorrhage there is no mention of the effects and the fate of extravasated blood. The author is impressed by the importance of tularemic pneumonia but not by that due to the Friedländer bacillus.

Some additional minor criticisms could be made. For example, while the illustrations on the whole are excellent, a few could probably be improved (figs. 115, 46, 92 and 369b). Figure 350 may well represent metaplasia rather than carcinoma. The primary lesion of anthrax is not a pustule, and this disease is contracted also from sheep and goats and their products. Table 4, page 163, is not truly representative of the systemic distribution of neoplasms.

There are numerous additional points on which issue might be taken with statements of fact or on the degree of emphasis. If there is any considerable demand for changes, they will, no doubt, be made by the author in his second edition.

The author of a textbook of pathology is confronted with several important decisions. Shall it be written primarily for medical students or at the reference and postgraduate level? Shall it be the work of one person or of a number of contributors? Shall it be confined to one volume? Dr. Moore has apparently attempted to compromise with regard to his audience. The book will probably prove to be more useful to the general student than to the professional pathologist because it often fails to give specific, pathognomonic details for diagnosis, so much desired by the latter. On the other hand, it does present for the pathologist many new points of view. Although the book was written by one author, he acknowledges the help of many consultants and associates. The book is in one volume. It is a heavy book of 1,338 pages. It might well be expanded to two volumes in future editions, with greater emphasis on general pathologic principles.

Here, then, is a new book which should find an important place for itself among textbooks of pathology. The author is to be congratulated, and students of pathology may consider themselves fortunate.

The Avitaminoses: The Chemical, Clinical and Pathological Aspects of the Vitamin Deficiency Diseases. By Walter H. Eddy, Ph.D., emeritus professor of physiological chemistry, Teachers College, Columbia University; and Gilbert Dalldorf, M.D., pathologist of the Grasslands and Northern Westchester Hospitals, Westchester County, New York. Third edition. Price $4.50. Pp. 438, with 47 illustrations. Baltimore: The Williams & Wilkins Company, 1944.

The first edition was published in 1937 and was reviewed in that year (ARCH. PATH. **24**:409, 1937). The present edition has nearly 100 more pages and 20 more illustrations.

Part I deals with: the chemical nature of the vitamins; vitamin behavior; vitamin requirements; the nature and the function of vitamin A, thiamine, riboflavin, nicotinic acid, pyridoxine, bios nutrilites and vitamins C, D, E and K. It contains many useful tabular summaries of vitamin units, dietary requirements, etc.

Part II presents in detail the etiologic factors, the experimental aspects, the clinical manifestations and the morphologic aspects of the human avitaminoses. In this part are 47 plates of, as a rule, excellent black and white illustrations of gross and microscopic appearances, nearly one half of which appear to be original. Plate 29 needs better description.

Part III describes methods of vitamin assay and tests for vitamin deficiency diseases.

The bibliography of parts I and II is arranged alphabetically according to chapters. The appendix contains a list of references to the estimation of vitamin potencies and two tables of the vitamin values of raw foods. The book contains a vast amount of information about vitamins and their relation to health and disease.

The Pathology of Internal Diseases. By William Boyd, M.D., LL.D., M.R.C.P. (Edin.), F.R.C.P. (Lond.), Dipl., Psych., F.R.C.S., professor of pathology and bacteriology in the University of Toronto, Toronto, Canada. Fourth edition, thoroughly revised. Pp. 857, with 374 illustrations. Price $10. Philadelphia: Lea & Febiger, 1944.

The first and second editions of this book were reviewed in earlier volumes of the ARCHIVES OF PATHOLOGY (**11**:682, 1931; **20**:965, 1935). There is no change in the general plan and scope of the book. As stated in the review of the second edition, the field covered is that "usually included under the term internal medicine except that acute infectious diseases, such as measles, scarlet fever, smallpox, tularemia, undulant fever and whooping cough, have not been included." It should be noted also that tropical diseases and diseases of the mouth, the pharynx, the esophagus, the vermiform appendix, the peritoneum, the sex organs and the skeleton are not considered. In the new edition several sections, notably those dealing with the cardiovascular system, have been largely rewritten, and much new material has been added. The book is well abreast of recent advances in its spheres of pathology. Of the new topics, alloxan diabetes and Rh factor have not found their way into the index. Four new colored plates and 22 new text figures have been added. The book is well illustrated. In the case of the colored plates the magnification is not given, and no mention is made of the stains in plates 1, 3 and 7. Within its field the book continues to be an excellent guide to the study of the pathology of internal diseases. The statements

on page 744 about the infectiousness of the patient with poliomyelitis and of the carrier of the virus of poliomyelitis are a rare instance of greater finality in the presentation than is warranted by present knowledge.

Lead Poisoning. By Abraham Cantarow, M.D., associate professor of medicine, Jefferson Medical College, and assistant physician and biochemist, Jefferson Hospital, Philadelphia; and Max Trumper, Ph.D., Lieutenant Commander, H-V(S), U.S.N.R., Naval Medical Research Institute, Bethesda, Md. Pp. 264. Price $3. Baltimore: The Williams & Wilkins Company, 1944.

The absorption, the transportation, the deposition and the excretion of lead form the topic of the first chapter. Then come discussions of the pathology and pathologic physiology of lead poisoning, its clinical manifestations and treatment. The principal part of the chapter on treatment consists of a description by May R. Mayers, M.D., of the industrial control of lead poisoning. There are chapters also on the occurrence of chronic plumbism; on the normal intake of lead; on lead in body fluids, blood and excretions; on lead products in industry (L. H. Schroeder, National Lead Company), and (by Morris B. Jacobs) on procedures for the determination of lead. There are many tabulations of useful data concerning lead and lead poisoning. There is an extensive bibliography, titles not given, arranged alphabetically according to authors' names. The book will be of interest to all medical men who are concerned with the prevention, the recognition and the treatment of lead poisoning.

Spina Bifida and Cranium Bifidum. Papers Reprinted from the New England Journal of Medicine with the Addition of a Comprehensive Bibliography, from the Department of Surgery of the Children's Hospital, Boston, and Harvard Medical School. By Franc D. Ingraham, M.D., assistant professor of surgery, Harvard Medical School; neurosurgeon, Children's Hospital; senior associate in neurosurgery, Peter Bent Brigham Hospital. Pp. 216, illustrated. Cambridge, Mass.: Harvard University Press, 1945.

The title well describes the contents. The articles discuss the results of observations of the malformations in question in 546 patients seen at the Children's Hospital, Boston, during the last twenty years. A survey of the cases is presented, including the surgical treatment. Three articles deal with occult spinal disorders, an unusual nasopharyngeal encephalocele, and the Arnold-Chiari malformation. The larger part of the book, 128 pages, contains the bibliography from 1556 to 1943, arranged by decades except for the early period. There is here a contribution of great value to those who are interested in the study and understanding of spina bifida and cranium bifidum.

Books Received

Report of the National Academy of Sciences. Fiscal Year 1942-1943. Pp. 165. Price 25 cents. Washington, D. C.: United States Government Printing Office, 1944.

Hospital for Joint Diseases, New York. Thirty-Seventh Annual Report—For the Year 1943. Pp. 102.

National Research Fellowships, 1919-1944. Administered by the National Research Council. Compiled by Neva E. Reynolds, recording secretary. Pp. 142. Washington, D. C.: National Research Council, 1944.

Archivos de la Sociedad Argentina de Anatomía Normal y Patológica. Dirigidos por D. Brachetto-Brian. Tomo V. Ano 1943. Pp. 143, illustrated. Buenos Aires: Asociación Médica Argentine, 1943.

Report of the Henry Phipps Institute of the University of Pennsylvania for the Period 1942-1943. Pp. 20.

Patients Have Families. Henry B. Richardson, M.D., associate professor of clinical medicine, Cornell University Medical College; attending physician, New York Hospital; visiting physician, Bellevue Hospital. Price $3. Pp. 408. New York: The Commonwealth Fund, 1945.